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Unexpected high species diversity among European stalked puffballs – a contribution to the phylogeny and taxonomy of the genus *Tulostoma* (Agaricales)

Mikael Jeppson[1], Alberto Altés[2], Gabriel Moreno[2], R. Henrik Nilsson[4,5], Yolanda Loarce[3], Alfredo de Bustos[3], Ellen Larsson[4,5]

1 *Lilla Håjumsgatan 4, SE-46135 Trollhättan, Sweden* **2** *Dpto. Ciencias de la Vida (Botánica), Universidad de Alcalá, E-28805 Alcalá de Henares (Madrid), Spain* **3** *Dpto. Biomedicina y Biotecnología (Genética), Universidad de Alcalá, E-28805 Alcalá de Henares (Madrid), Spain* **4** *University of Gothenburg, Biological and Environmental Science, Box 461, SE-40530 Göteborg, Sweden* **5** *Gothenburg Global Biodiversity Centre, Box 461, SE-405 30 Göteborg, Sweden*

Corresponding author: *Ellen Larsson* (ellen.larsson@bioenv.gu.se)

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Abstract

A three-gene data set was generated to explore species diversity and delimitations within the stalked puffballs (*Tulostoma*, Agaricales) in Europe. Data on species from other parts of the world were included for comparison of species concepts and distribution ranges. Sequence data from 26 type specimens are included. The phylogenetic analyses support *Tulostoma* as monophyletic. Eleven major clades, 37 minor clades, and 20 single branches were recovered and found to correspond to 30 described species and 27 species without scientific names.

Five species are here described as new to science: *Tulostoma calcareum*, *T. calongei*, *T. eckbladii*, *T. grandisporum*, and *T. pannonicum*. In total we report 26 described, and 19 undescribed, species from Europe. An epitype for *T. fimbriatum* with ITS sequence data is selected to fix the name.

The recovered tree topology was not in congruence with the current infrageneric classification of *Tulostoma*, suggesting that many of the morphological characters used for segregation of taxa are plesiomorphic or homoplasious. Spore ornamentation and hyphal structure of the peridium are found to be reliable characters for delimitation of species.

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The majority of the species occur in the dry, arid areas of southern and east central Europe but a few are shown to be restricted to humid temperate regions in the North. The study confirms that species with smooth or sub-smooth spores are restricted to dry and arid habitats whereas species with more strongly ornamented spores occur in humid habitats.

Areas with steppe vegetation in Hungary and Spain are here identified as hot spots for *Tulostoma* species diversity.

Key words

Gasteroid fungi, molecular systematics, species diversity hot spot, steppe vegetation, taxonomy

Introduction

The genus *Tulostoma* Pers. : Pers. was erected by Persoon (1794, 1801) to accommodate puffball species with a "peridium pedicellatum", opening with an "ore cylindraceo cartilagineo". Persoon included two species in the genus, *T. brumale* and *T. squamosum*. The former had previously been illustrated by Tournefort (1700) as *Lycoperdon parisiense* and later described by Linnaeus (1753) as *Lycoperdon pedunculatum*. Fries (1829) mentioned four species, viz. *T. mammosum* (including *T. squamosum*), *T. fimbriatum*, *T. tortuosum* and *T. laceratum* (=*Schizostoma lacerata*). However, in a later work, Fries (1849) replaced *Tulostoma* with *Tulasnodea*, honouring the Tulasne brothers, contemporary mycologists based in Paris. However, according to the rules of nomenclature, *Tulostoma* must be considered the correct name for this characteristic genus, being sanctioned by Persoon (1801). An orthographic variant, *Tylostoma*, is sometimes seen in older literature. Early contributions to the knowledge of *Tulostoma* are the works of Bottomley (1948), Coker and Couch (1928), Cunningham (1925, 1932), Hollós (1904, 1913), Lloyd (1906), Long (1944, 1946), Long and Ahmad (1947), Morgan (1891), Petri (1904, 1909), Pouzar (1958), Shvartsman and Filimonova (1970), Smith (1951), and White (1901).

Schroeter (1876) studied young and old basidiomata of *Tulostoma* and described the development of the basidiomata, in particular basidia and capillitium. Later Wright (1955) surveyed important morphological characters in the genus. This was followed by the world monograph of *Tulostoma* (Wright 1987), where he accepted 139 species. Wright's species concept was largely based on type studies and studies of numerous additional herbarium specimens. With the introduction of scanning electron microscopy (SEM), detailed spore morphology could be used as a base for taxonomic rearrangements and introduction of new species (Altés and Moreno 1995, Altés and Moreno 1999, Altés et al. 1999, Moreno et al. 1992a, Moreno et al. 1997).

From the late 20th century the arid regions of Mexico have been intensely explored with regard to *Tulostoma* (Esqueda et al. 2004, Hernández-Navarro et al. 2015, Moreno et al. 1995b, Piña et al. 2010, Wright et al. 1972). In addition, South America, particularly Brazil, has recently been subject to detailed studies on *Tulostoma* (e.g. Baseia and Milanez 2002, Cortez et al. 2009, Silva et al. 2007). Silva's (2006) thesis on *Tulostoma* substantially increased the knowledge of the genus in Brazil. For South Africa, Coetzee

(2007) published a key to the known taxa and Moreno et al. (1995a), Moreno et al. 2002, and Kreisel and Scholler (2008) added species for the continent. For Asia, Liu (1979), Asai (2004), Asai and Asai (2008), Dörfelt and Bumžaa (1986), Hussain et al. (2016), Sesli et al. (2000), and others have published survey reports on the occurrence of the genus. In Europe, numerous papers dealing with the genus have been published: Altés et al. (1996), Antonín and Kreisel (2008), Calonge (1992, 1998), Calonge et al. (2007), Calonge and Martín (1992), Jeppson (2006a,b, 2008), Kreisel (2004), Kříž (2011), Martín (1990), Martín and Calonge (1994), Moreno et al. (1990), Mrazek et al. (1995), Sarasini (2003, 2005), Schubert and Specht (2013a,b), Specht and Schubert (2012), Specht et al. (2016), and Tomaszewska et al. (2011).

The introduction of molecular data has led to major changes in fungal systematics and taxonomy. Hibbett et al. (1997) were the first to show that the puffball genus *Tulostoma* belongs in Agaricales. Although the genus has been intensively studied from a morphological point of view, so far comparatively few molecular phylogenetic studies with a focus on *Tulostoma* have been published, and these are mainly geographically restricted and deal with a low number of species (cfr. Hernández-Caffot et al. 2011, Hussain et al. 2016).

In the present study we use a three-gene phylogeny with the aim to explore species diversity and phylogenetic relationships within *Tulostoma*. The data set includes the ITS, LSU, and Tef-α regions of the majority of the European species. In addition, species described from other parts of the world are included for comparison of species concepts and distribution ranges. Retrieval of ITS sequence data from type and other herbarium specimens was attempted when permission for sequencing was given and as the condition of the material allowed.

Material and methods

Morphological methods

The genus *Tulostoma* is easy to identify to genus level in the field. Precise determination to species level is, however, often difficult, because the characters used are few in number and subtle in nature. According to Index Fungorum (http://www.indexfungorum.org; accessed July 1, 2016) there are well over a hundred *Tulostoma* names to consider, many of them known from old and scanty type materials at best. Our sampling and selection of species was compiled through field studies and collecting in various parts of Europe during the last 25 years (collections stored in GB and AH).

Additional collections and type specimens were studied as loans from herbaria around the world (BPI, BRA, C, FH, L, LD, LISU, MA, NY, O, PC, PRM, S, TRH, and UPS). Herbarium acronyms are in accordance with Index Herbariorum (http://sweetgum.nybg.org/science/ih/). Species very recently recorded from Europe (*T. bruchi, T. leslei, T. palatinum,* and *T. vulgare*) are not included in this study (Antonín and Kreisel 2008, Calonge 1992, 1998, Specht et al. 2016).

Collections were mostly photographed in situ, dried, and later studied in the laboratory under a stereo-microscope. Morphological features are named in accordance with Wright (1987). For microscopic studies, samples of mature gleba were mounted in Cotton blue + lactophenol and heated to boiling. Features of the peridium were studied in Melzer's reagent. A light microscope equipped with a Dino-Eye Eyepiece Camera was used for studies of micro-morphological characters. Spores were measured at a magnification of 1000x, using the Dino-Lite 2.0 software (www.dino-lite.eu). A minimum of 20 spores were measured for each sample. All spore measurements are given excluding ornamentation. Studies under SEM were conducted according to the procedure of Moreno et al. (1995b).

Molecular methods

Taxon sampling

For this study, 183 ingroup specimens were sequenced, including the type specimen of 24 species. ITS sequence data of the type specimens of *T. domingueziae* (HQ667597) and *T. ahmadii* (KP738712) were taken from GenBank (Hernández-Caffot et al. 2011, Clark et al. 2016). Specimens were selected with the aim to cover all the species known to occur in Europe and from a broad geographic distribution within the area. In addition, specimens from North America and Central Asia were included for comparison of species concepts and distribution ranges. The sequenced specimens shown in Figure 1a–c are indicated with an asterisk in the lists of specimens examined and are listed with GenBank accession numbers as Suppl. material 1.

Three additional ITS and LSU sequences from Hernández-Caffot et al. (2011) were retrieved from GenBank and included in the data set (HQ667594, HQ667598-HQ667599). Based on results from earlier molecular phylogenetic studies of Agaricales (Hibbett et al. 1997, Matheny et al. 2006), *Lycoperdaceae*, *Cyathus, Crucibulum,* and *Inocybe* were selected as outgroup taxa.

DNA extraction, PCR, and sequencing

Sequence data from three genetic markers were generated for the study: the complete ITS region and about 1400 bases of the 5′end of the nuclear ribosomal LSU DNA, and about 1000 bases of translation elongation factor subunit 1 alpha (Tef-1a). DNA extractions were performed using DNeasy Plant Mini kit (Qiagen, Hilden, Germany), PCR reactions, and sequencing were performed as described in Larsson and Örstadius (2008). Primers used to amplify the complete ITS region and the 5′end of the LSU region were ITS1F (Gardes and Bruns 1993) and LR21, LR0R, and LR7 (Hopple and Vilgalys 1999). For Tef-1a we used EF983F and EF2218R (Rehner and Buckley 2005). Primers used for sequencing were ITS1, ITS4 (White et al. 1990), LR0R, LR5,

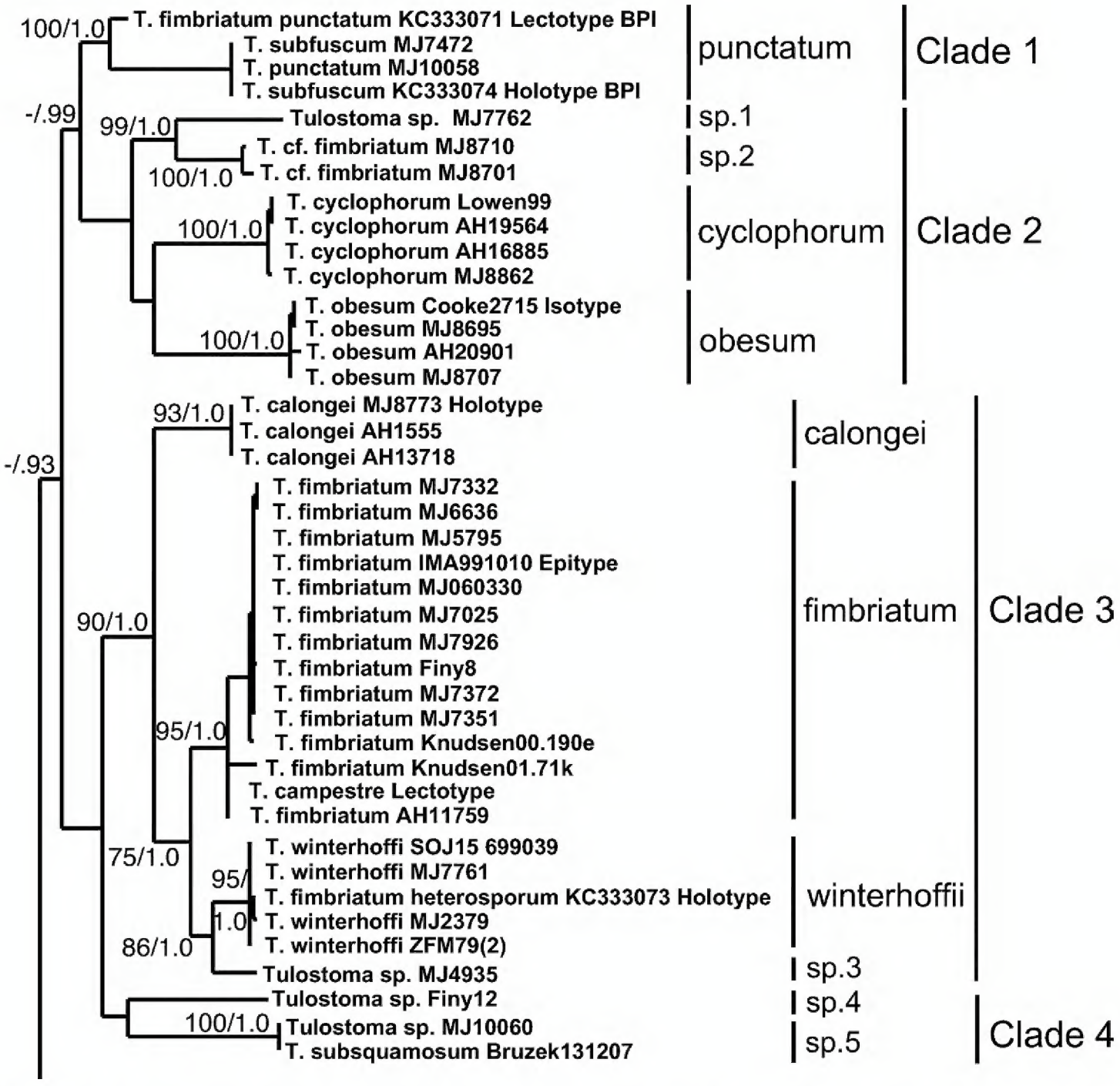

Figure 1. a–c One of the most parsimonious trees obtained from the parsimony analysis based on ITS, LSU, and Tef-1α sequence data of *Tulostoma*, with focus on species diversity in Europe. Bootstrap values and Bayesian Posterior Probabilities are indicated on branches. The major clades (1-11) are marked with a scale bar. The clades represent species or species groups that are discussed in more detail in the text.

and LR3R (Hopple and Vilgalys 1999, Tedersoo et al. 2015), EF983F, and EF2218R. Type specimens were extracted using a modified CTAB method, and PCR and sequencing followed protocols described in Larsson and Jacobsson (2004).

Phylogenetic analyses

Sequences were edited and assembled using Sequencher 5.1 (Gene Codes, Ann Arbor). Alignment of individual genes was performed using the L-INS-i strategy as implemented in MAFFT v. 7.299 (Katoh and Standley 2013). The alignments were adjusted manually using AliView (Larsson 2014). Sequences generated for this study have been

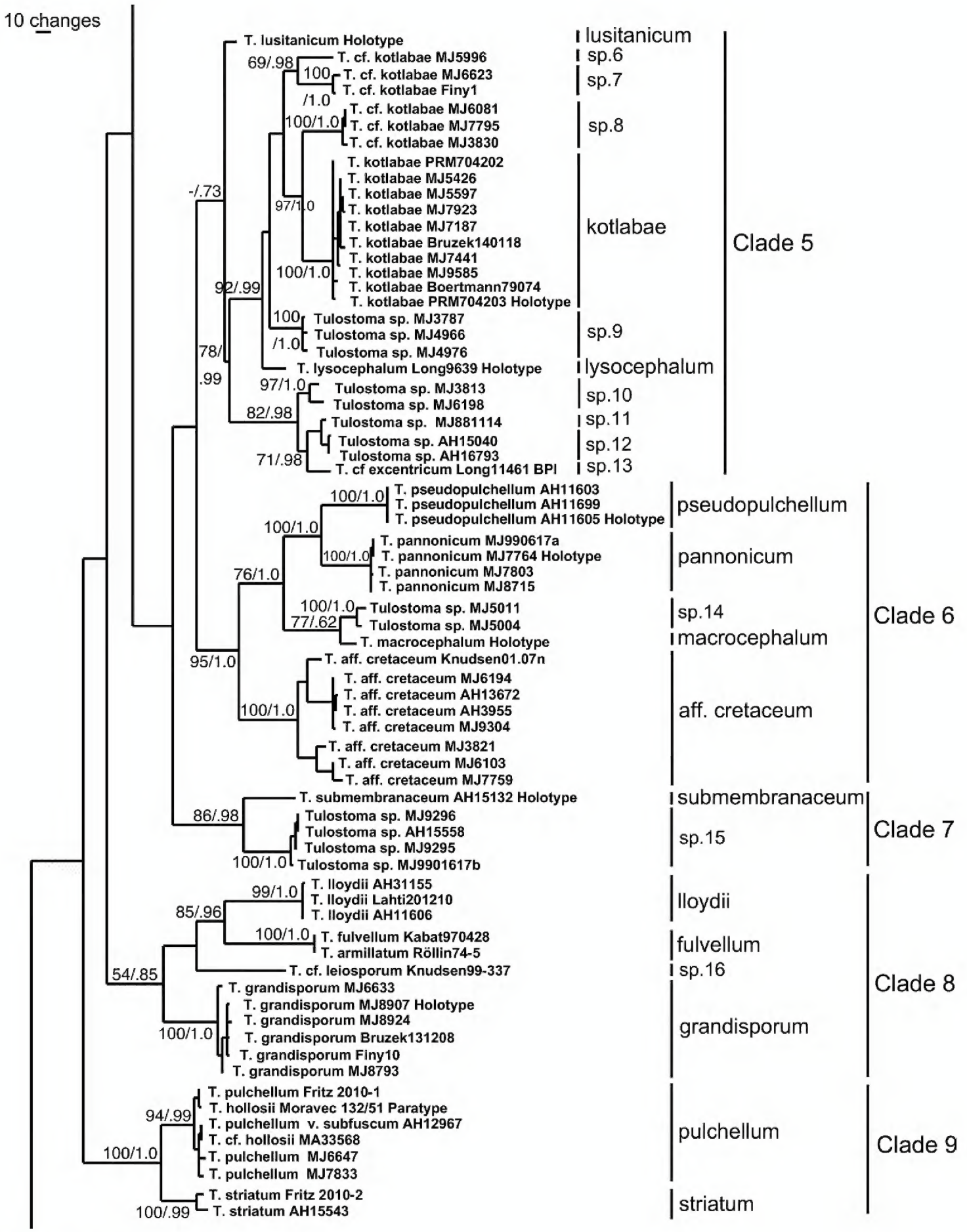

Figure 1. Continue.

deposited in GenBank (ITS/LSU: KC333055–KC333076, KU518952–KU519107, KX513825–KX513827, KX576541–KX576549, KX583648, KX640979-KX640988; Tef-1(: KU843875–KU844001).

Separate phylogenetic analysis was done for all three genetic markers to test for overall congruity of the phylogenetic signal. The trees were found to be compatible with

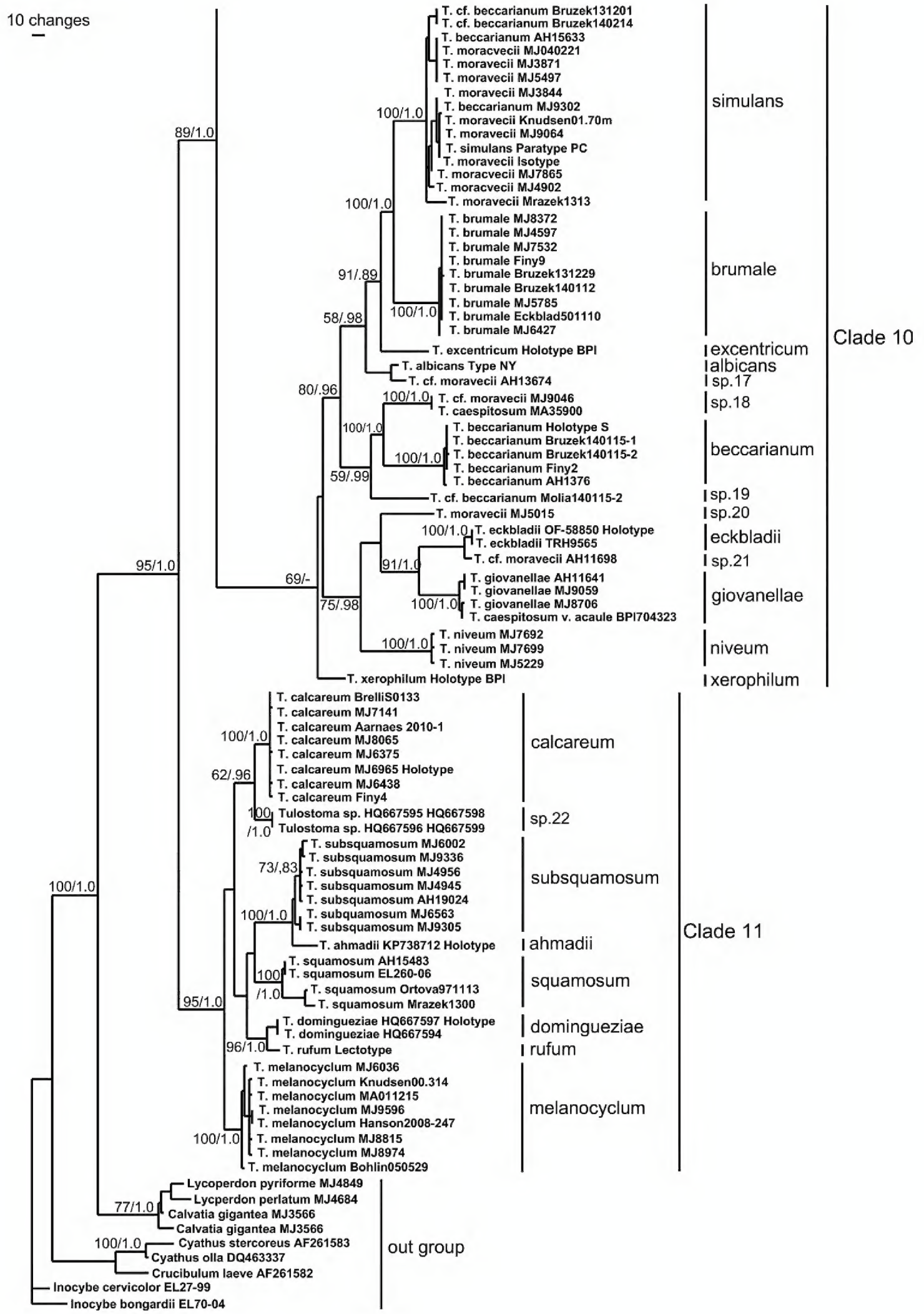

Figure 1. Continue.

respect to the overall clades (results not shown), and the three genetic markers were concatenetad for the final analyses. Heuristic searches for the most parsimonious trees were performed using PAUP*. All transformations were considered unordered and equally weighted. Variable regions with ambiguous alignment, mainly from the ITS region, were excluded, and gaps were treated as missing data. Heuristic searches were performed with 1,000 random-addition sequence replicates, TBR branch swapping, and the MulTrees option in effect. Relative robustness of clades was assessed by the bootstrap method using 1,000 heuristic search replicates with 100 random taxon addition sequence replicates and TBR branch swapping, the latter saving at most 25 trees in each replicate.

Bayesian phylogenetic analyses were carried out in MrBayes 3.2.6 (Ronquist et al. 2012), with a best-fit model of nucleotide evolution for the separate gene partitions supplied by MrModeltest 2.2 (Nylander 2004). Eight default-setting Metropolis-Coupled Markov Chain Monte Carlo (MCMCMC) chains were run for 10 million generations with trees sampled every 5,000 generations and an initial burn-in of 50%. After discarding the trees prior to the burn-in threshold, a 50% majority-rule consensus phylogram was computed from the remaining 1,000 trees.

Results

Molecular analysis

The aligned complete dataset, including sequences downloaded from GenBank, consisted of 198 taxa and 3,340 characters. After exclusion of ambiguous regions, mainly from the ITS1 and ITS2, 3,005 characters remained for the analysis. Of these, 1,992 were constant, 226 were variable but parsimony uninformative, and 787 (26%) were parsimony informative.

The maximum parsimony analysis yielded 3625 equally parsimonious trees (length=3,726 steps, CI= 0.3814, and RI= 0.8228). One of the trees is presented as a phylogram in Figure 1a–c.

The bootstrap analysis recovered *Tulostoma* as monophyletic with 95 % bootstrap support (BS). Thirty-seven minor clades and 20 single branches were recovered, corresponding to 30 described species and 27 without a scientific name. Five of these clades are here described as new species to science. Eleven major clades, ranging from virtually unsupported to having strong support, within the ingroup are recognized and named as Clades 1-11 in the phylogenetic tree (Figure 1a–c). These clades are further described and discussed below.

As suggested by MrModeltest, the nucleotide evolution model HKY+G was used for the ITS1 spacer; SYM was used for the 5.8S gene; HKY+G was used for the ITS2 spacer, and GTR+I+G were used for the nLSU and TEF genes in the Bayesian analysis. The MCMC analysis converged well in advance of the burn-in threshold and chain mixing was found to be satisfactory, as assessed by using Tracer v1.5 (Drummond et al. 2012). Also in the Bayesian analysis, *Tulostoma* was recovered as monophyletic with

strong support (a Bayesian posterior probability (BPP) of 1.00). The Bayesian tree topology is similar to the MP bootstrap tree. The same clades and single branches recovered in the maximum parsimony analysis were also recovered in the Bayesian analysis, with the minor differences that several clades with low or no bootstrap support received a moderate to high BPP value. BPP values are indicated on the corresponding branches in Figure 1a–c.

None of the newly generated ITS sequences were significantly similar to the "most wanted fungi" ITS dataset of Nilsson et al. (2016) as explored through BLAST.

Comments on the major clades

Clade 1 (100, 1.0) includes only one species, *T. punctatum*. The exoperidium is hyphal and the mouth is fibrillose-fimbriate. It is similar to *T. fimbriatum* in macro-morphology but has smaller and more coarsely ornamented spores without anastomosing ridges. The species was treated as a variety of *T. fimbriatum* by Wright (1987).

Clade 2 (unsupported) includes four species that all have a hyphal exoperidium. Two of the species (sp.1, sp. 2) are unidentified. The specimens of these originate from halophytic vegetation in Spain and sand steppe vegetation in Hungary, respectively. Both have a fibrillose-fimbriate mouth, thus inviting comparison to the widespread *T. fimbriatum* in morphology. *Tulostoma cyclophorum* has a typically fibrillose-fimbriate mammose mouth but differs from other European species of *Tulostoma* by having abundant mycosclereids, i.e. rounded to elongated cells on the endoperidial surface, and subreticulate spores. *Tulostoma obesum* has a smooth endoperidial surface that lacks mycosclereids and the spores are completely smooth.

Clade 3 (90, 1.0) includes four species with similar morphology. They all have a hyphal exoperidium, fibrillose-fimbriate mouth and rather stout basidiomata, features that are characteristic of *T. fimbriatum*. An ITS sequence from the lectotype of *T. campestre* (=*T. fimbriatum* var. *campestre*) was generated and forms a monophyletic clade with the sequences of *T. fimbriatum* sensu stricto. As the morphology also is in congruence, we regard it as a synonym. An epitype is selected for *T. fimbriatum* (see taxonomy section) from the same district in Sweden as the holotype to fix the name also by ITS sequence data (Nilsson et al. 2012, Ariyawansa et al. 2014). *Tulostoma winterhoffii*, a recently described species from Germany, is identical with the ITS sequence from the holotype of *T. fimbriatum* var. *heterosporum*, and thus replaces that name. We describe a new species from Spain, *T. calongei*. It is similar to *T. fimbriatum* in habit but differs in spore morphology and molecular data. The unidentified *T.* sp. 3, also with the habit of *T. fimbriatum*, collected in Hungary, indicates the occurrence of cryptic speciation within this morphologically similar group. Although having been considered synonymous with *T. fimbriatum* or mere varieties of it (Altés and Moreno 1995, Wright 1987), ITS data (not shown in the tree) of type materials of the extra-European taxa *T. egranulosum, T. readeri,* and *T. tuberculatum* indicate them to be distinct species. The *T. fimbriatum* complex is in need of further studies.

Clade 4 (unsupported) includes two unidentified species (*Tulostoma* spp. 4 and 5) collected in sandy habitats in Central Europe. *Tulostoma* sp. 4 is fairly similar to *T. fimbriatum* in habit. *Tulostoma* sp. 5 has a circular, tubularly protruding mouth, a hyphal exoperidium, and irregularly ornamented, verrucose-echinate spores. The two species are very different morphologically and their placements in the tree are ambiguous and without significant support.

Clade 5 (-, .73) is an unsupported clade including 11 species. The holotype of *T. lusitanicum* (Figure 12), a species recently (2000) described from Portugal, is placed on a single branch basal in the clade, but without significant support. However, the residual subclades form a moderately supported clade (78/.99, Figure 1b). ITS sequence data of the holotypes of *T. kotlabae, T. lysocephalum* (from North America), and *T. lusitanicum* were included, but still we have eight unidentified clades (*Tulostoma* spp. 6-13). This indicates a high and previously unrecognized species diversity within the group, including cryptic speciation. The clade is in need of further study, and the addition of more sequence data appear to be needed to resolve the phylogenetic relationships. *Tulostoma kotlabae* and spp. 6-13 share morphological characters such as the circular, more or less tubularly protruding mouth, a hyphal exoperidium, pale colours of the basidiomata, and weakly to moderately ornamented spores. *Tulostoma* sp. 12 is known only from the Mediterranean area, and an Italian finding (AH 16793) was published as *T. kotlabae* by Altés et al. (1994). *Tulostoma lysocephalum* differs by having a fibrillose-fimbriate mouth (Figure 22).

Clade 6 (95, 1.0) includes five species. *Tulostoma* aff. *cretaceum* forms a strongly supported clade that comprises specimens from Russia, Hungary, and Spain. In morphology they are characterized by stout basidiomata with pale colours and a hyphal or slightly membraneous exoperidium, a very dark, chocolate brown colour of the mature gleba, an indistinct, irregular mouth that with age becomes lacerate, combined with totally smooth spores. However, as there is substantial sequence variation that correlates with geographic distribution within the clade (Russia, Hungary and Spain, respectively), it must be regarded as a complex of species. More data such as the sequence of the type of *T. cretaceum* are needed to resolve species delimitation within the *Tulostoma* aff. *cretaceum* clade. Sequence data from the holotype of *T. macrocephalum* from North America show this species to be closely related to *Tulostoma* sp. 14, which includes two specimens from Spain, collected in a halophytic habitat. Both species have a hyphal exoperidium but differ in basidiomata size and spore wall ornamentation. *Tulostoma pseudopulchellum,* a species characterized by a membranous exoperidium, a fibrillose-fimbriate mouth, and finely and irregularly ornamented spores comes out as a sister clade with strong support to what we describe as a new species (*T. pannonicum*, see taxonomy section). The new species is recognized by having small smooth spores and has previously been reported from Hungary as *T. leiosporum* (cfr. Jeppson et al. 2011, Rimóczi et al. 2011).

Clade 7 (86, .98) includes two species. *Tulostoma submembranaceum* was described from Mexico by Moreno et al. (1995b). The exoperidium is thinly membranous-verrucose, the mouth is fibrillose-fimbriate, and the spores have low verrucae that have a tendency to form crests. *Tulostoma* sp. 15 from Hungary and Spain, more or less

matches the macro-morphology of *T. submembranaceum* but differs in spore ornamentation (irregularly rugulose) and molecular data.

Clade 8 (54, .85) includes four species. Uniting morphological characters are the presence of a fimbriate-fibrillose mouth and totally smooth spores. Two well-known species are *T. fulvellum* and *T. lloydii*. One of the recovered clades represents a species with large spores, characteristic of sandy habitats of East Central Europe. It is proposed as a new species (*T. grandisporum*, see taxonomy section). *Tulostoma* sp. 16 is a specimen from Siberia that in macro-morphology is similar to the newly described *T. grandisporum* but has significantly smaller spores. Both have irregular, undulating, or ragged inner walls of the capillitium. This striking character is also present in the lectotype of *T. leiosporum*, from which we were not able to obtain sequence data.

Clade 9 (100, 1.0) includes *T. pulchellum* and *T. striatum* and corresponds to section *Poculata* Pouzar & Moravec. The exoperidium is distinctly membranous, detaching in flakes. The endoperidium is white, velvety in young specimens, and has a mammose, fibrillose-fimbriate mouth that sometimes is surrounded by a more or less delimited peristome. They share macro-morphological features but can be readily separated by their spores that are finely asperulate in *T. pulchellum* and distinctly striate in *T. striatum*.

Clade 10 (69, -) includes 14 species. All species in this clade have a mouth that is circular and more or less tubular, or conically protruding. The spores are moderately ornamented. *Tulostoma simulans*, a species described from N. America, is according to our results widely distributed also in Europe. In some parts of Europe, *T. simulans* has been mistaken for *T. beccarianum* and the species *T. moravecii.* The latter now appears to be a later synonym. The well-known and widely distributed *T. brumale* is closely related, but clearly distinct from *T. simulans*, both in molecular and morphological data. *Tulostoma beccarianum* is a rare species described from Italy by Bresadola, but with recent records from Spain and East Central Europe. *Tulostoma* spp. 17-21, all represented by few specimens, are similar in morphology to *T. beccarianum* and *T. simulans,* and represent un-named cryptic species that need further attention. The ITS of the types of *T. albicans, T. excentricum,* and *T. xerophilum,* three species from North America, were generated and included, but did not match with any of our unknown species, and the latter occurs on a single basal branch in Clade 10. *Tulostoma giovanellae* is mainly associated with sandy, and often halophytic, habitats in southern and central Europe. The sister clade to *T. giovanellae* represents a species from northernmost Scandinavia that we describe as a new species (*T. eckbladii*, see taxonomy section). Within the clade there are also *T.* sp. 21, an unidentified Spanish specimen, closely related to *T. eckbladii,* and the characteristic bryophilous *T. niveum.*

Clade 11 (95, 1.0) includes eight species that morphologically are characterized by having rather dark colours of the stipe, endoperidia with initially brownish colours, and moderately to strongly ornamented, verrucose-echinate spores. The widely distributed *T. squamosum* has a membranous-verrucose exoperidium of sphaerocysts, which upon maturity forms a dark reticulum on the brownish endoperidium. The closely related *T. subsquamosum* has a paler, hyphal exoperidium with only scattered sphaerocysts, and never forms a reticulum. It was described from India and reported from Spain by Altés et al. (1996) and is here confirmed also from East Central Europe (Hungary

and Slovakia). Sequence data indicate a close relationship with the recently described *T. ahmadii* (Hussain et al. 2016). *Tulostoma melanocyclum* is a widely distributed species in Europe. The recently described *T. domingueziae*, reported from South America (Hernández-Caffot et al. 2011), as well as *T. rufum* (lectotype; North America) come out as sister species. One of the recovered clades is described as a new species (*T. calcareum,* see taxonomy section) that has a wide European distribution. The sequence data of *T.* sp. 22 represent an undescribed species from South America.

Taxonomy

Recognized European species in *Tulostoma* and the descriptions of five new species

***Tulostoma beccarianum* Bres., in Petri, Ann. Mycol. 2(5): 413. 1904.**
Figure 2a

Holotype. ITALY, Pisa: Beccari (S!)*.

The holotype material of *T. beccarianum,* described in 1904, was sequenced in this study. Identical sequences were obtained from recently collected material from Hungary, Slovakia, and Spain. The species was given a detailed description by Altés and Moreno (1993) based on the holotype. The newly collected samples have rather large and stout basidiomata, spore-sacs measuring up to 22 mm in diameter, and a stem reaching 120 mm. The exoperidium is indistinct or hyphal, and the endoperidium is smooth, dirty white-greyish, with a circular, shortly raised mouth. The stem base is widened and forms a volva-like structure. The capillitial septa are slightly widened. The capillitium seems to break up easily at the septa, leaving segments with somewhat widened, rounded ends. The type material shows irregular, undulating inner walls of the capillitium, a character that is also noted in the recently collected material. The spores are irregularly verrucose, 4–5 µm, av. 4.5 µm, ornamentation excluded (Figure 2a) and agree with those of the holotype (4.7–6 µm, ornamentation included; Altés and Moreno 1993). The synonymisation of *T. beccarianum* with *T. simulans*, as proposed by Altés and Moreno (1993), was contradicted by the molecular analyses. The name *T. beccarianum* has apparently sometimes been misapplied for *T. simulans* (syn. *T. moravecii*). *Tulostoma* sp. 19 includes a single collection from Cyprus, with more or less identical morphology. It is currently treated as an undescribed species until more material becomes available.

Habitat and distribution. In semi-shaded to exposed localities in dry grasslands (Hungary and Slovakia) and halophytic vegetation on sand (Spain). Apparently a very rare species.

Other specimens examined. HUNGARY, **Bács-Kiskun**: Kiskunhalas, 19 Jan. 2014, *P. Finy* 2 (GB)*. SLOVAKIA, **Košice**: Borša, 15 Jan. 2014, M. Lazon, P. Brůžek 140115-1 (GB)*; Ibidem, P. Brůžek 140115-2 (GB)*. SPAIN, **Castellón**: Oropesa del Mar, bajo *Artemisia gallica*, 30 Sept. 1976, G. Moreno (AH 1376)*; Cabanes, arenas marítimas (albufera) con *Artemisia* sp. y musgos, 10 Nov. 1990, G. Moreno, A. Burguete (AH *12953*).

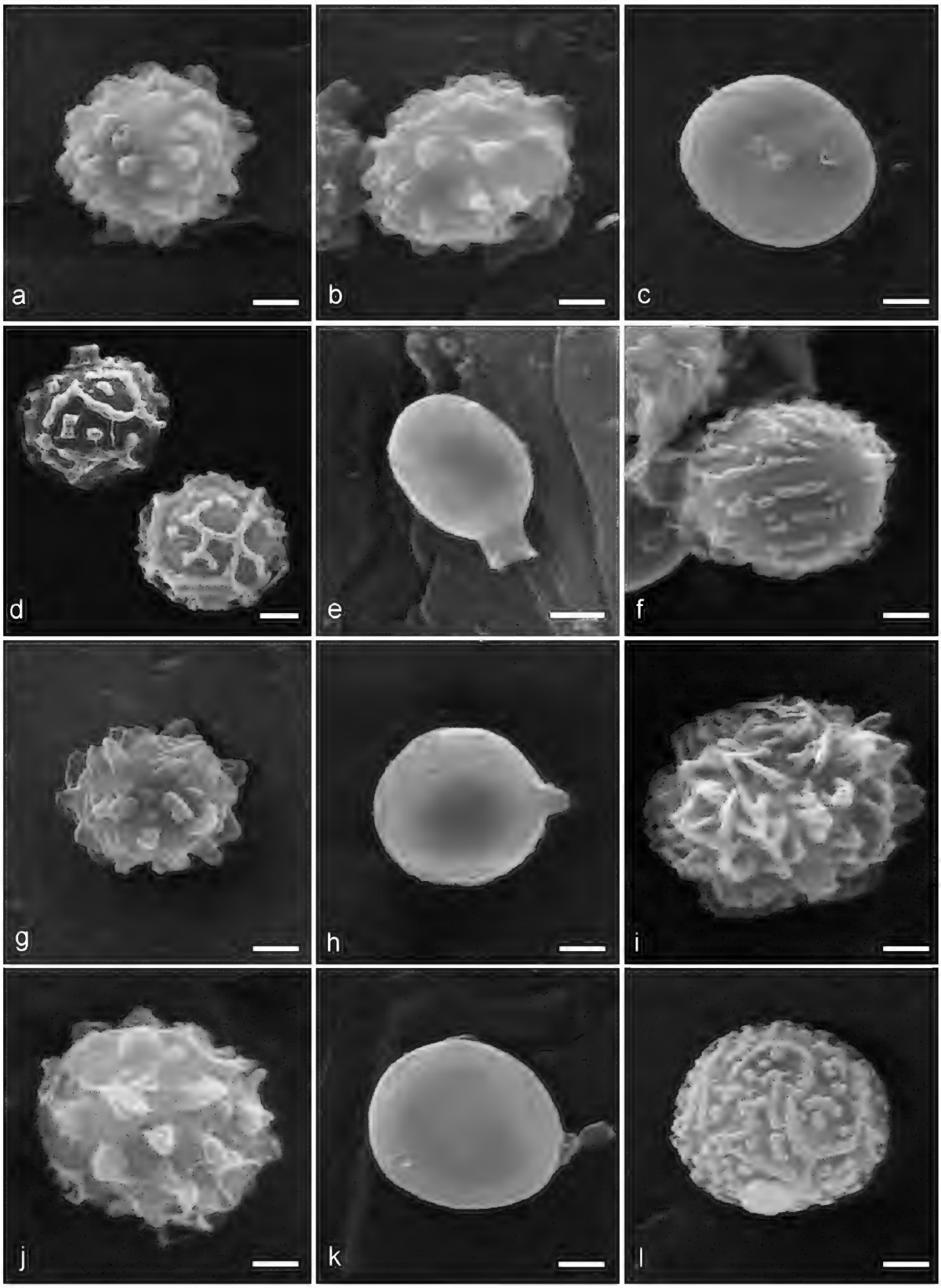

Figure 2. Spore ornamentation under SEM of European species: **a** *T. beccarianum* (Finy 2, GB) **b** *T. brumale* (M. Jeppson 8372, GB) **c** *T.* aff. *cretaceum* (M. Jeppson 3821, GB) **d** *T. cyclophorum* (AH 16885) **e** *T. fulvellum* (AH 13415) **f** *T. giovanellae* (AH 11641) **g** *T. kotlabae* (M. Jeppson 5597, GB) **h** *T. lloydii* (AH 11606) **i** *T. melanocyclum* (S. Hanson 2008-247, GB) **j** *T. niveum* (M. Jeppson 7699, GB) **k** *T. obesum* (AH 20901) **l** *T. pulchellum* (M. Jeppson 7833, GB). Scale bars: 1 µm.

***Tulostoma brumale* Pers., Neues Mag. Bot. 1: 86. 1794.**
Figure 2b

Type. FRANCE, "circa Parisios" (L); several collections by Persoon available at L, but no type seems to have been formally designated (cfr. Wright 1987).

Tulostoma brumale is well characterized by its membranous exoperidium, the circular-tubular mouth –which is usually surrounded by a brown peristome – and its abrupt widenings of the capillitial septa. SEM-photos of the spores show an irregular ornamentation of blunt, broad-based verrucae (Figure 2b). It can be separated from *T. simulans* by the exoperidial features, its slightly smaller spores, and the presence of irregular crystals adhering to the capillitial walls.

Habitat and distribution. According to Wright (1987), *T. brumale* is recorded from N and S America and Asia (Georgia). Shvartsman and Filimonova (1970) reported it from Central Asia and Rebriev and Gorbunova (2007) added findings from Siberia. In Europe it is one of the more common species of *Tulostoma*, recorded north to 60° in Fennoscandia. It occurs in all types of sandy and calcareous grasslands, sand dunes, and sand steppe vegetation, as well as on moss covered rocks and stone walls. It is often associated with mosses, particularly *Syntrichia* spp.

Specimens examined. CZECH REPUBLIC, **S. Moravia**, Břeclav, Pavlov, Děvín-Kotel-Soutéska NPR, 29 Dec. 2013, P. Brůžek 131229 (GB)*; **Praha**: Lochkovské údolí-Cikánka, limestone, pebble, gravel, 12 Jan. 2014, Brůžek 140112 (GB)*. FRANCE, **La Vendée**: Saint Juan, Plage de la Tonelle, among *Syntrichia* on sand dune, 25 Jan. 2007, J. Jeppson, M. Jeppson 8372 (GB)*. HUNGARY, **Pest**: Tatárszentgyörgy, 11 Jan. 2011, P. Finy 9 (GB)*. NORWAY, **Vest-Agder**: Lista, Bausjø, øst for Tjørve, på mose på sanddyne, 10 Nov. 1950, F.-E. Eckblad 501110 (O 58849)*. SLOVAKIA, **Stražovské vrchy**: Ďurďové, calcareous grassland, 6 Oct. 2005, T. Knutsson, J. Jeppson, M. Jeppson 7532 (GB)*. SWEDEN, **Gotland**: Gotska Sandön, gräshed med *Corynephoretum* nära stranden, 24 May 2013, T. Knutsson 2013-134 (GB); **Öland**: Sandby, Sandby kyrkvägskäl, sand steppe vegetation on road verge, 15 Nov. 2003, M. Jeppson 6427 (GB)*; **Skåne**: Åhus, Horna bränneri, among *Syntrichia* in sand steppe vegetation, 24 Oct. 1998, M. Jeppson 4597 (GB)*; Köpinge, Fritorpet, 26 Oct. 2001, M. Jeppson 5785 (GB)*.

***Tulostoma calcareum* Jeppson, Altés, G. Moreno & E. Larss., sp. nov.**
MycoBank: MB819399
GenBank: KU519086 (ITS-LSU), KU843881 (Tef-1a)
Figure 3

Holotype. Sweden, Södermanland, Mörkö, Egelsvik, Kalkberget, on calcareous soil under *Artemisia campestris* in rocky slope, 5 Nov. 2004, M. Jeppson 6965 (GB!, isotype AH).

Etymology. The name refers to its habitat requirement, on calcareous sandy soil or among calcareous rocks and cliffs.

Description. Spore-sac subglobose, often somewhat depressed, 5–12 mm. Exoperidium hyphal-verrucose, deciduous, sometimes persisting as whitish verrucae scattered

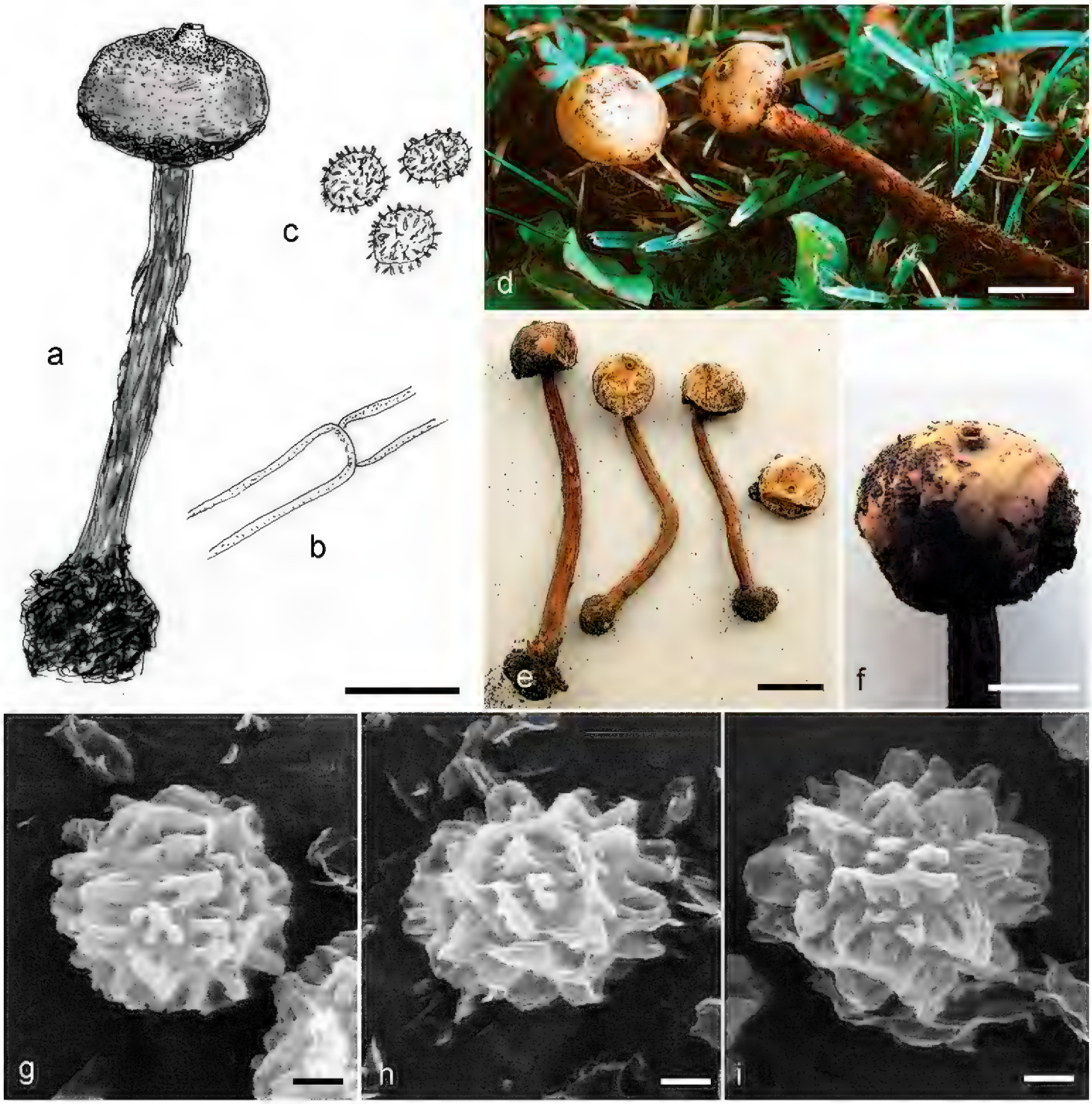

Figure 3. *T. calcareum*, holotype (M. Jeppson 6965, GB): **a, e** basidiocarps **b** capillitium **c, g–i** spores **f** detail of spore-sac showing the tubular mouth **d** basidiocarps (M. Jeppson 6513, GB). Scale bars: **a, d–e** = 10 mm; **b–c** =10 µm; **f** = 5 mm; **g–i** = 1 µm.

on the endoperidial surface. Endoperidium brownish-ochraceous, initially rather dark coloured, with age fading to greyish white. Mouth circular, shortly tubular (Figure 3f), surrounded by a greyish or brownish peristome. Socket separated from the stem, inconspicuous. Stem slender, 20–50 x 2–3 mm, initially orange brown, with age warm reddish brown, longitudinally furrowed, smooth to fissured and appressed scaly, with a basal mycelial bulb (Figure 3a,e). Gleba ochraceous to ferrugineous brown. Capillitium 4–10 µm with medium-thick walls, sometimes abundantly ramified. Septa not or very slightly widened (Figure 3b). Spores subglobose, 4.0–6.0 µm (av. 4.7–5.0 µm), verrucose-echinate. SEM-photos show spines in groups with connected tips, sometimes coalescing to form ridges and crests (Figure 3g–i).

Habitat and distribution. Occurs in dry, exposed to semi-shaded situations in calcareous, sandy habitats and on calcareous rocks and cliffs. It is currently on record from Hungary, Norway, Spain, and Sweden.

Notes. *Tulostoma calcareum* was commonly misinterpreted as *T. squamosum* (cfr. Nitare 1997), under which name it is red-listed as Critically endangered (CR) in Sweden (Artdatabanken 2015). It differs from *T. squamosum* by having a hyphal to slightly verrucose exoperidium, by lacking sphaerocysts, and by never forming the reticulate pattern typical of *T. squamosum*. The stipe is also less squamulose. Spores and capillitial characters are almost identical. It differs from *T. melanocyclum*, with which it can easily be confused, by having a more robust stature, more orange-reddish brown colours of the stipe, and slightly smaller spores (av. 4.7–5.0 µm vs 5.2–5.4 µm in *T. melanocyclum*). SEM-photos of *T. melanocyclum* (Figure 2i) show similar spore ornamentation but with spines with less tendency to coalesce and form ridges. In macro-morphology *T. calcareum* is fairly similar to *T. ferrugineum* D.M. Oliver & Hosford, a species described from northwestern USA, whose type material no one has been able to locate (Wright 1987). However, according to its original description, it differs from *T. calcareum* by having distinctly widened capillitial septa.

Specimens examined. HUNGARY, **Budapest**: Pilisszántó, on dolomite grassland, 31 Dec. 2013, P. Finy 4, (GB)*. NORWAY, **Oppland**: Vågå, 15 Jul. 2010, A. Breili S0133 (GB)*. SPAIN, **Girona**: Lloret de Mar, Platja de Fenals, sandy grassland with scattered pines, near hotel buildings, Oct. 2010, J-O. Aarnaes 2010-1 (GB)*. SWEDEN, **Södermanland**: Mörkö, Egelsvik, Kalkberget, on calcareous soil under *Artemisia campestris* in rocky slope, 5 Nov. 2004, M. Jeppson 6966 (GB). **Öland**: Böda, Ramsnäs, Hagudden, torrmark mellan fritidshus och strandvallen, 5 Apr. 2007, E. Hultqvist, T. Knutsson, U. Andersson, T. Gunnarsson, M. Jeppson 9077 (GB); Gårdby, sandgräshed, 6 Mar. 1999, K. Bringer (GB); Gårdby, V. om hembygdsgården, sand steppe vegetation, 25 Sept. 2004, J. Jeppson, M. Jeppson 7141 (GB)*; Högsrum, Karums alvar, dry calcareous grassland around *Juniperus* bush, 11 Nov. 2006, Ölands Botaniska Förening, M. Jeppson 8065 (GB)*; Hulterstad, Alby, på Ancylusvallen vid infarten till skolan, väg-/trädgårdskant i gräsmatta med mossor på sandig jord, 18 Apr. 1997, H. Lundqvist, T. Knutsson 97-034 (GB); Hulterstad, Alby skola, i gräsmatta och rabatt, 15 Oct. 2002, S. Jeppson, J. Jeppson, M. Jeppson 6513 (GB); Hulterstad, Triberga, sand steppe vegetation, 15 Nov. 2003, J. Jeppson, M. Jeppson 6375 (GB)*; Ibidem, 15 Nov. 2003, M. Jeppson 6438 (GB)*; Norra Möckleby, Dörby, torräng i kanten av den forna järnvägsbanken, 4 Apr. 2009, E. Hultqvist, T. Knutsson, U. Andersson, T. Gunnarsson, M. Jeppson 9084 (GB); Norra Möckleby, Bettorps vägskäl, sandstäppsvegetation, 21 Jul. 2006, T. Knutsson, U. Andersson, T. Gunnarsson, M. Jeppson 8777 (GB); Resmo, "Resmokorridoren", 5 May 2005, U. Andersson, T. Gunnarsson (GB); Räpplinge, Borgholms slottsalvar, kalkhällmark i ängshavretorräng, 11 Nov. 2001, J. Jeppson, M. Jeppson 5743 (GB); Ibidem, M. Jeppson 5744 (GB); Sandby, Sandby kyrkvägskäl, sandy roadside, 24 Sept. 2004, J. Jeppson, M. Jeppson 7121 (GB).

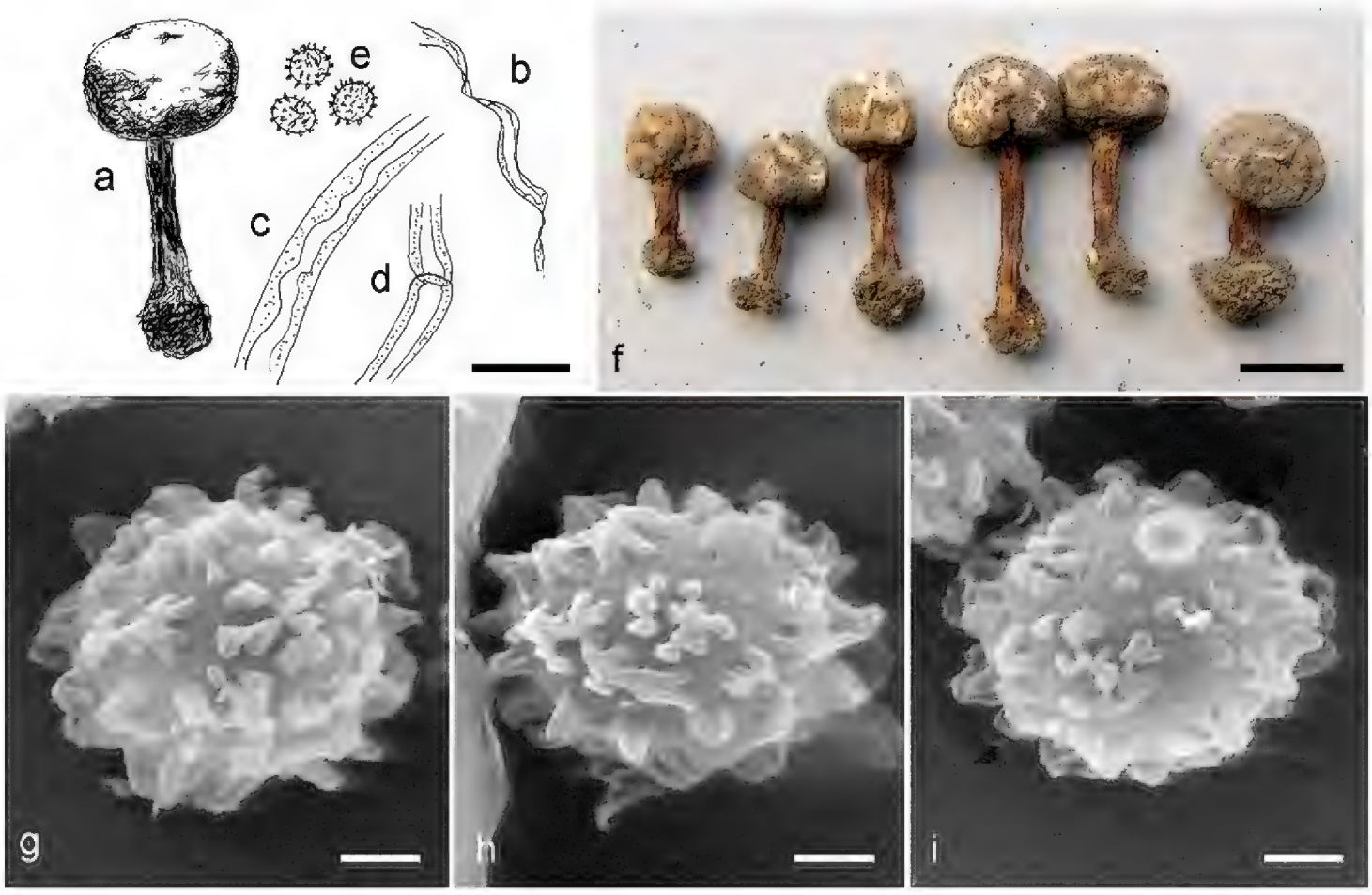

Figure 4. *T. calongei*, holotype (M. Jeppson 8773, GB): **a, f** basidiocarps **b–d** capillitium **e, g–i** spores. Scale bars: **a, f** = 10 mm; **b–e** = 10 µm; **g–i** = 1 µm.

Tulostoma calongei **Jeppson, Altés, G. Moreno & E. Larss., sp. nov.**
MycoBank: MB819400
GenBank: KU518973 (ITS-LSU), KU844000 (Tef-1a)
Figure 4

Holotype. Spain, Madrid, Casa de Campo, in exposed situation on dry sandy soil, S. Jeppson, J. Jeppson, M. Jeppson, 21 Nov. 1989, M. Jeppson 8773 (GB!, isotype AH).

Etymology. Named in honour of the Spanish mycologist Francisco D. Calonge.

Description. Spore-sac 8–10 mm, subglobose. Exoperidium granulose hyphal, encrusting soil, sometimes detaching in small flakes, whitish-greyish, rather persistent (Figure 4f). Endoperidium smooth, whitish. Mouth fimbriate. Stipe rather short, 15–20 x 1.5–2.5 mm, warm reddish brown, with darker appressed scales and a basal mycelial bulb (Figure 4f). Gleba ochraceous to ferrugineous brown. Capillitium 3–6 µm, thick-walled, with uneven inner walls (Figure 4c-d). Some ribbon-like hyphae present (Figure 4b). Septa rare, slightly widened. Spores globose-subglobose, 3.5–4.5 µm (av 4.1 µm) ornamentation excluded, verrucose-echinate, with spines frequently fused at their tips forming little developed pyramidal groups (Figure 4g-i).

Habitat and distribution. *Tulostoma calongei* has been collected only from central Spain, usually on sandy acidic soils.

Notes. Although usually having a less robust stature, this new species is very similar in macro-morphology to *T. fimbriatum*, with which it has been confused. The

main differences are that the spores are slightly smaller and have a conical-pyramidal ornamentation, without the characteristic crests of *T. fimbriatum* (Figure 7c–e). The molecular data also clearly support them as distinct but closely related species, with *T. calongei* occuring as a sister clade to *T. fimbriatum* (Clade 3, Figure 1a). The distribution area for the species is likely to increase after reexamination of additional samples identified as *T. fimbriatum* from Meridional Europe.

Other specimens examined. SPAIN, **Madrid**, El Pardo, suelo arenoso, 12 Nov. 1978, E. Álvarez (AH 13718)*; Manzanares el Real, La Pedriza, jaral en suelo arenoso, 21 Dec. 1977, C. Ladó (AH 1555)*; Carabanchel Alto, camino arenoso junto a pradera, Nov. 1989, A. Martínez (AH 12586, GenBank KC333064); Ibidem, Nov. 1990, A. Martínez (AH 12686, GenBank KC333065).

Tulostoma aff. *cretaceum* Long, Mycologia 36(4): 321. 1944.

Figures 2c, 5a–b

Remarks. Clade 6 in our study encompasses some closely related taxa previously attributed to *T. volvulatum* I.G. Borshch. and *T. obesum* Cooke & Ellis (cfr. Hollós 1904, Altés et al. 1999, Rimóczi et al. 2011). The main characteristics are the rather large basidiomata with hyphal exoperidium and almost pure white endoperidium with an indistinct to fibrillose-fimbriate mouth (or irregular scar) that with age becomes lacerate. The stipe base is provided with a volva and a prominent pseudorhiza. The spores are smooth and the capillitium is fragile, breaking up in segments with rounded ends. In our material this complex is represented by collections from Hungary, Russia (Altay Republic), and Spain. They are recovered as closely related and can be considered as cryptic species with a strong geographical signal. In morphology they fit the original description of the American species *T. cretaceum* fairly well. The Spanish collections deviate from the Hungarian specimens in having a stouter habit.

Habitats and distribution. *Tulostoma cretaceum* was originally described from New Mexico (USA) and later reported from Argentina (Wright 1987) and Baja California, Mexico (Moreno et al 1995b). There are recent records from Mexico (Sonora) and Brazil (Esqueda et al. 2004, Silva et al. 2007). In Europe *T.* aff. *cretaceum* is found in sand steppe vegetation in Central Hungary and in calcareous steppe habitats in Spain.

Specimens examined. HUNGARY, **Bács-Kiskun**: Fülöpháza, Fülöpházai Homokbuckák, in open sand in sand steppe vegetation, 13 Jun. 1999, J. Jeppson, M. Jeppson 4901 (GB): Ibidem, 11 Apr. 2006, T. Knutsson, T. Gunnarsson, J. Jeppson, M. Jeppson 7759 (GB)*; Keleshalom, Keleshalmi Borokás, in open sand in sand steppe vegetation, 18 Jun. 1997, S. Jeppson, J. Jeppson, M. Jeppson 3821 (GB)*; **Pest**: Örkény, former military training field, sand steppe vegetation, in open sand, 5 Nov. 2001, J. Jeppson, M. Jeppson 6103 (GB)*. RUSSIAN FEDERATION, **Altay**: Chuya River/Kuektanar River, 31 km W of Kosh-Agach on M52, 21 Aug. 2001, H. Knudsen 01.70n (a), (mixed collection; C)*. SPAIN, **Ciudad Real**: Parque Nac. Tablas de Daimiel, sobre terreno básico, 30 Apr. 1983, J.M. Barrasa (AH 3955,

Figure 5. Habit of a selection of studied collections: **a** *T.* aff. *cretaceum* (M. Jeppson 7759, GB) **b** *T.* aff. *cretaceum* (M. Jeppson 6194, GB) **c** *T. obesum* (M. Jeppson 8695, GB) **d** *T. punctatum* (M. Jeppson 10058, GB) **e** *T. simulans* (M. Jeppson 7865, GB) **f** *T. simulans* (M. Jeppson 9302, GB) **g** *T. subsquamosum* (M. Jepsson 9336, GB) **h** *T. winterhoffii* (M. Jeppson 2379, GB). Scale bars: 10 mm.

GB)*; **Huelva**, La Palma del Condado, calcareous pasture with *Thymus* sp., 1 Jan. 2003, S. Jeppson, J. Jeppson, M. Jeppson 6194 (GB)*; **Madrid**, Alcalá de Henares, cerros de Alcalá, gypsophilous steppe vegetation, 3 Mar. 2011, G. Moreno, A. Altés, A. Stridvall, E. Larsson, M. Jeppson 9304 (GB)*; Alcalá de Henares, Cerro Malvecino, terreno margoso con algo de yeso, 7 Nov. 1990, equipo micología UAH (AH 13672, GB)*.

***Tulostoma cyclophorum* Lloyd, Mycol. Writings 7: 25. 1906.**
Figure 2d

Holotype. SOUTH AFRICA, Stoneman, herb. Lloyd 4495 (BPI).

Tulostoma cyclophorum is a morphologically characteristic species, originally described from South Africa, with a tomentose, white to ochraceous endoperidium covered with mycosclereids and a distinctly membranous exoperidium. The mouth is silky fibrillose-fimbriate. Its spores are unique among European *Tulostoma* lineages, being more or less reticulate (visible under SEM, Figure 2d; asperulate under light microscope).

Habitat and distribution. Mostly found in semi-shaded localities among grasses and herbs on lawns in city parks on somewhat sandy soil. From Europe currently reported from France, Italy, Hungary, and Spain (Demoulin 1984, Jeppson et al. 2011, Moreno et al. 1990, Sarasini 2005). It appears to have a cosmopolitan world distribution.

Specimens examined. ARGENTINA, **Catamarca**: Dpto. Capayán, Concepción, picada a Pomán, en vegetación arbustiva del "Chaco serrano" a orillas de un camino, 23 May 1998, M.J. Silverio-Reyes (AH 19564, GB)*. FRANCE, **Auvergne**: Saint-Priest-en-Murat, 14 Oct. 1986, Priou 86150 (AH 13418). HUNGARY, **Szatmár-Bereg**: Nyíregyháza, College of Nyíregyháza, Stadion út, urban lawn, under Platanus, 21 Sept. 2009, M. Jeppson 8862 (GB)*. SPAIN, **Huelva**: Aracena, 5 Nov. 2001, L. Romero de la Osa (AH 28451); **Madrid**: Alameda de Osuna, suelo arenoso bajo *Pinus pinea*, 22 Oct. 1994, V. González (AH 16885, GB)*; Parque del Oeste, in grass, 20 Oct. 1984, R. Lowen 99 (GB)*.

Tulostoma eckbladii Jeppson, Altés, G. Moreno & E. Larss., sp. nov.

MycoBank: MB819401
GenBank: KU519068 (ITS-LSU), KU843951 (Tef-1a).
Figure 6

Holotype. Norway, Nordland, Saltdal, Junkerdalsura, på sten blandt mos, 4 Sept. 1957, I. Jørstad (O F-58850!).

Etymology. Named in honour of the Norwegian mycologist Finn-Egil Eckblad (1923-2000).

Description. Spore-sac subglobose to onion-shaped, 6–8 mm in diameter. Exoperidium not observed. Endoperidium smooth, initially brownish, later ochraceous white. Mouth circular, shortly tubular, with a more or less brown peristome. Socket inconspicuous. Stem stout, 25–30 x 3–4 mm, flattened, light brown, somewhat longitudinally furrowed, almost smooth to finely appressed scaly, attenuated towards the base, with whitish basal rhizomorphs attached to moss rhizomes (Figure 6d). Mature gleba ochraceous-ferrugineous. Capillitium 4–6 µm with rather thick walls, with scattered septa that are slightly widened (Figure 6b). Spores subglobose, 4.5–5.5 µm (av. 4.9 µm) ornamentation excluded, coarsely but not very densely verrucose. SEM photos show isolated, irregular verrucae with broad bases (Figure 6e–g).

Habitat and distribution. *Tulostoma eckbladii* is so far only known from two findings in northern Norway, in both cases growing among mosses on calcareous boulders.

Notes. The species reminds of *T. niveum* in its choice of habitat on moss-covered rocks. It can, however, be distinguished by its stouter basidiomata and the presence of a brown peristome. Whether or not the flattened stipe is a constant species character cannot be decided from the available material. The spores are similar, as observed from SEM photos (Figure 2j, 6e-g). It is distinguished from *T. brumale* by the presence of a stout stipe, the less widened capillitial septa, and the larger spores. Also *T. simulans* is morphologically similar but is recognized by its more slender basidiomata and different habitat and distribution. The two findings of *T. eckbladii*

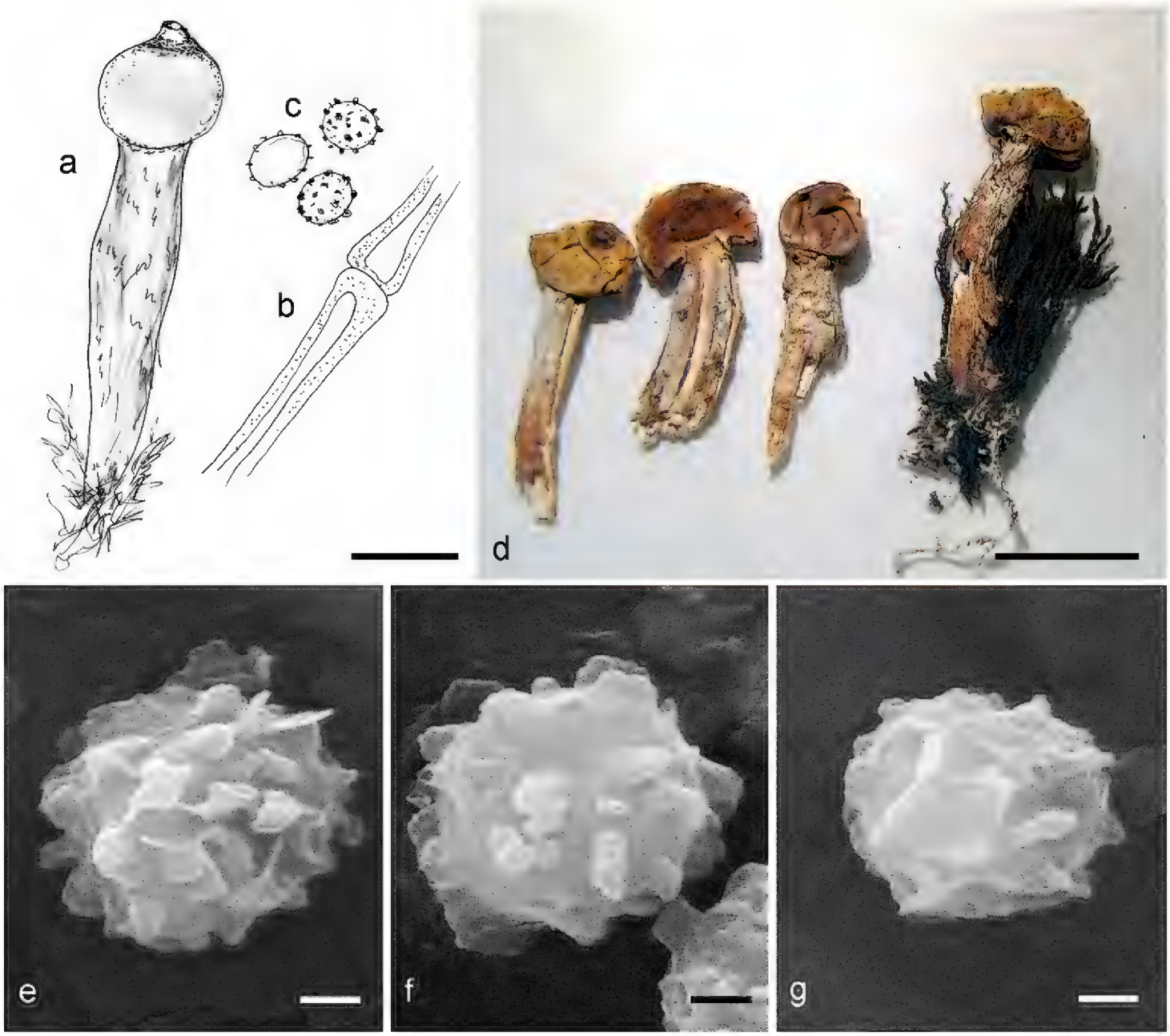

Figure 6. *T. eckbladii*, holotype (O F-58850): **a, d** basidiocarps **b** capillitium **c, e–g** spores. Scale bars: **a, d** = 10 mm; **b–c** = 10 µm; **e–g** = 1 µm.

constitute northern extremities in the distribution of the genus *Tulostoma*. It has previously been reported and discussed by Brochmann et al. (1981) as *Tulostoma* sp.

Other specimens examined. NORWAY, **Finnmark**: Porsanger, Hestnes, 17 Jul. 1993, B. Jonsell (as *T. kotlabae*, TRH 9565)*.

Tulostoma fimbriatum **Fr., Syst. Mycol. 3(1): 43. 1829.**
Figures 7, 8

≡ *Tulostoma fimbriatum* var. *campestre* (Morgan) G. Moreno, An. Jard. bot. Madr. 36: 18 (1980) [1979].

≡ *Tulostoma campestre* Morgan, J. Cincinnati Soc. Nat. Hist. 12: 163. 1889. Lectotype: USA, Nebraska, Webber (NY!)*.

≡ *Tulostoma fibrillosum* V.S. White, Bull. Torrey Bot. Club 28: 433. 1901. Holotype. Canada, East shore of Lake Huron, Sept. 1891, J. Dearness (NY!).

Holotype. Sweden, Skåne, Lomma, ca 1820, E. Fries (UPS F-175998!). Epitype (designated here, MycoBank typification number: MBT374178): Sweden, Skåne, Ravlunda, Klammersbäck, sandy grassland under *Pinus*, 10 Oct. 1999, I. Månsson 991010 (GB!)*.

GenBank: KU518963 (ITS-LSU), KU843904 (Tef-1α).

Tulostoma fimbriatum is one of the more frequently recorded species of the genus. It was described by Fries (1829) from southernmost Sweden. In the Elias Fries Herbarium at UPS there is a collection from the type locality (Lomma, S. Sweden; Fries 1829, 1849). On the label there is the handwriting of T. M. Fries (Elias Fries' son) but the material is likely to be an authentic collection of Elias Fries. The material is scanty (one basidiome glued to a piece of cardboard) with the endoperidial mouth zone destroyed and with indistinct micro-morphological characters. Nevertheless it should be considered a holotype, but since its characteristics are ambiguous, an epitype is here chosen from recently collected and sequenced material originating from southernmost Sweden. *Tulostoma fimbriatum* can be distinguished from other species with similar macro-morphology by its spores, which have a low ornamentation of verrucae coalescing in ridges and Y-shaped crests (Figure 7c–e). The lectotype of *T. campestre* (=*T. fimbriatum* var. *campestre*) forms a monophyletic clade with *T. fimbriatum* sensu stricto. As the morphology (Figure 8) also is congruent we regard them as synonymous. See also *T. winterhoffii*, with which this species has previously been confused.

Habitat and distribution. In dry and exposed habitats; both on calcareous and acidic soils, widespread in Europe reaching 60 'N in Scandinavia. Cosmopolitan distribution but in Europe often confused with the widely distributed *T. winterhoffii*.

Other specimens examined. GERMANY, **Hamburg**: Klein Nordende, Wasserstrasse 49, vägkant i allé, 3 Feb. 2016, H. Lundqvist (GB). HUNGARY, **Bács-Kiskun**: Lakitelek, Szikra, at Railway station, in ruderal sand steppe vegetation, 17 Feb. 2004, J. Jeppson, M. Jeppson 6636 (GB)*; Tolna, Németkér, 2 Nov. 2012, P. Finy 8, (GB)*. RUSSIAN FEDERATION, **Altay**: Chuya River, 21 Aug. 2001, H. Knudsen 01.71k (C)*; **Irkutskaya obl.**: Lake Baykal, Cape Rytyo, 8 Sept. 2000, H. Knudsen 00.190e (C)*. SLOVAKIA, **Podunajská nížina**: Marcelová, Marcelovské piesky, among grasses on sand dune, 16 Oct. 2004, M. Jeppson 7332 (GB)*; **Záhorská nížina**: Lakšárska Nová ves, Dolina, on sandy road verge in *Pinus* forest, 17 Nov. 2014, M. Jeppson 7351 (GB)*; Plavecký Mikuláš, in dry sandy soil (*Corynephoretum*) close to sand pit, 22 Jun. 2006, M. Jeppson 7926 (GB)*; Sväty Ján, Sekule, sandy grassland, 19 Oct. 2004, M. Jeppson 7372 (GB)*. SPAIN, **Cádiz**: Pinar de Barbate, on sandy roadside in *Pinus pinea* woodland, 28 Dec. 2004, S. Jeppson, J. Jeppson, M. Jeppson 7025 (GB)*; **Cáceres**: La Bazagona, inmediaciones del P. N. Monfragüe, en pradera ácida arenosa con *Geastrum campestre*, 18 Jan. 1990, G. Moreno, A. Altés (AH 11759)*; **Castellón**: Oropesa del Mar, playa Morro-gos, bajo *Artemisia gallica*, 1 Oct. 1975, J. Gómez (AH 1350, GenBank KC333055; AH 1465, GenBank KC333056); **Girona**: Port de la Selva, 8 Dec. 1997, O. Llimona, (ex BCC-16, AH 25191, GenBank KC333068); **Madrid**: Alcalá de Henares, cerros de Alcalá, bajo *Pinus halepensis*, Dec. 1988, J. Álvarez, J.A. Esteban, C. Illana (AH 11493, GenBank KC333059); Ibidem, 1 Mar. 1991, Curso

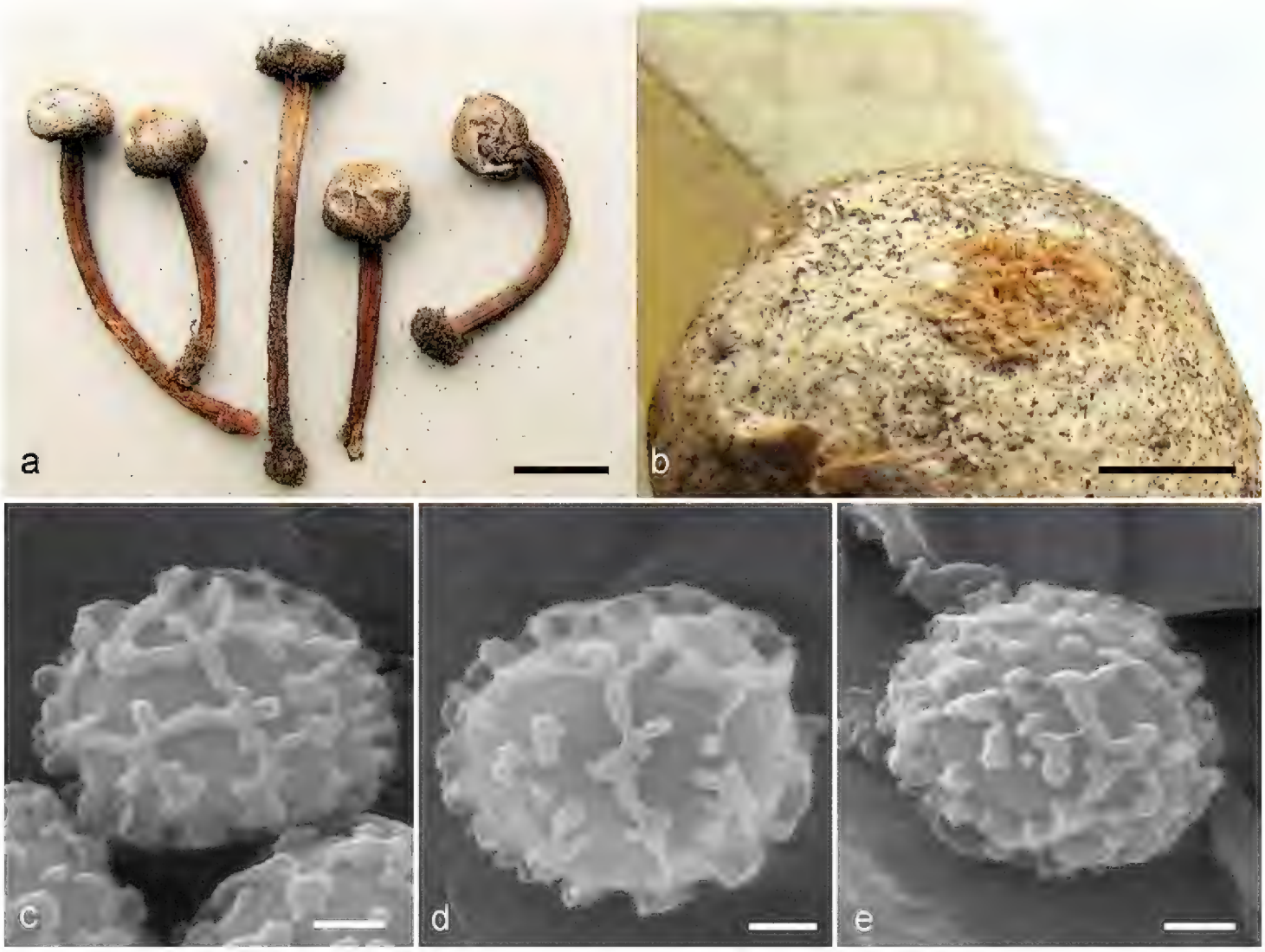

Figure 7. *T. fimbriatum*, epitype (GB): **a** basidiocarps **b** detail of spore sac showing the fibrillose-fimbriate mouth **c–e** spores. Scale bars: **a** = 10 mm; **b** = 2 mm; **c–e** = 1 µm

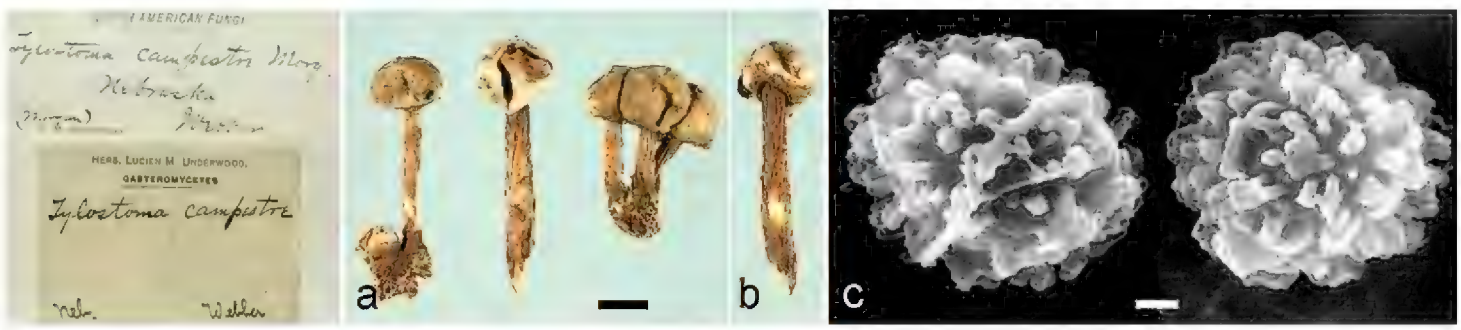

Figure 8. *T. campestre*, lectotype (NY): **a–b** basidiocarps **c** spore ornamentation under SEM. Scale bars: **a, b** = 10 mm; **c** = 1 µm.

doctorado gasteromicetos 1991 (AH 13114, GenBank KC333066); Carabanchel, terreno básico, 25 Nov. 1989, V. González, C. Ortiz, A. Martínez (AH 11757, GenBank KC333062); **Salamanca**: Béjar, en suelo removido de huerta, con Urtica, 6 Jan. 2000, J. Giménez (AH 25238, GenBank KC333069); **Valladolid**: Peñafiel, bajo *Pinus*, 7 Jan. 1982, P. Asenjo (AH 2973, GenBank KC333058). SWEDEN, **Blekinge**: Sölvesborg, Tredenborg, 30 Mar. 2006, B. Nilsson, M. Jeppson 060330 (GB)*; **Skåne**: Ravlunda, Haväng, Lindgrens ängar, sandstäpp, 27 Oct. 2001, S. Hanson, M. Jeppson 5795 (GB)*.

Tulostoma fulvellum **Bres., in Petri, Ann. Mycol. 2(5): 425. 1904.**
Figure 2e

≡ *Tulostoma armillatum* Bres., in Petri, Ann. Mycol. 2(5): 422. 1904. Holotype. ITALY, Rovereto, 1897, P. Pl. Giovanella (S!).

Holotype. ITALY, Trento: "juxta vias", Bresadola (K).

Our sequence data confirm *T. fulvellum* as a well-defined species in Clade 8, as a sister species to *T. lloydii*. In agreement with Wright (1987), we consider *T. fulvellum* to be the valid name for this species.

Habitat and distribution. *Tulostoma fulvellum* is a species of humus-rich habitats in shaded to semi-shaded habitats. Reported from Europe (France, Germany, Italy, Slovakia, Spain, and Switzerland; Bataille 1910, Candoussau 1973, Jeppson 2008, Kabát 1987, Monthoux and Röllin 1974, Wright 1987) and Japan (Asai and Asai 2008).

Specimens examined. SLOVAKIA, **Bratislava**: 28 Apr. 1997, V. Kabát 970428 (BRA)*. SPAIN, **Vitoria**, en serrín, 28 Oct. 1972 (ex herb. Candoussau 3551, AH 13414). – SWITZERLAND, **Genève**: 18 Feb. 1974, O. Röllin 74-5 (pers. herb. P. Specht)*; Moulin-de-Vert, suelo calizo con hojarasca, 27 Jun. 1974, O. Monthoux 4264 (AH 13415).

Tulostoma giovanellae **Bres., Fungi Trident. 1(1): 63. 1881.**
Figure 2f

≡ *Tulostoma volvulatum* I.G. Borshch., Zap. Imp. Akad. Nauk 7: 189. 1865. Holotype. TURKMENISTAN: in arenosis deserti Kara-Kum, Oct. 1857, I.G. Borshschov (LE 1938).

Holotype. ITALY, Trento: in Horto Cappucinarum, 1880, Bresadola (S!)

Our sequence data confirm *T. giovanellae* as a well-defined species. Morphologically it is characterized by its finely verrucose spores with verrucae arranged in radial lines, as seen under SEM (Figure 2f). Some photos of the holotype collection at S (habit and spore ornamentation under SEM) were included in Altés et al. (1999). Since *T. volvulatum* is an older synonym, it takes priority over *T. giovanellae*. Art. 57 of the Melbourne Code allows an exception to the priority rule, preventing the use of a name that incurs in serious conflict with the taxonomic concept traditionally associated with it. Also, Art. 56 allows rejection of a name in that circumstance. A formal proposition will be put forward to the General Committee (to be published in Taxon) that the name *T. volvulatum* become a *nomen rejiciendum*, whereas *T. giovanellae* would be a conserved name to be used in its traditional sense.

A sequence from a collection labelled *T. caespitosum* var. *acaule*, originating from Patouillard's herbarium (BPI), proved to be identical with *T. giovanellae*. See discussion (below) for issues around interpretation of *T. caespitosum* and *T. caespitosum* var. *acaule*.

Habitat and distribution. Africa, Asia, and Europe. It is a typical Mediterranean species found on sandy or halophytic soils in more or less exposed sites, but it is also recorded from anthropogenic habitats in Austria, Hungary, and Germany (Bohus and Babos 1977, Kreisel 1984, Rimóczi et al. 2011).

Other specimens examined. AFRICA, N. Patouillard 4639 (as *T. caespitosum* var. *acaule*, herb. Lloyd 15426, BPI 704323)*. HUNGARY, **Bács-Kiskun**: Kecskemét, 1 Dec. 1898, L. Hollós (S). SPAIN, **Alicante**: Punta del Sabinar, en dunas costeras, sobre restos enterrados de Posidonia oceanica, 28 May 1989, C. Bartolomé, J. Álvarez (AH 11641, GB)*; **Almería**: Tabernas, Puente de los Callejones, in halophilous vegetation on sandy rambla, 22 Apr. 2008, M. Jeppson 8706 (GB)*; **Zaragoza**: Candasnos SW, in halophilous steppe vegetation on road verge, 7 Apr. 2009, E. Larsson, U. Andersson, T. Gunnarsson, T. Knutsson, M. Jeppson 9059 (GB)*.

***Tulostoma grandisporum* Jeppson, Altés, G. Moreno & E. Larss., sp. nov.**
MycoBank MB819402
GenBank: KU519003 (ITS-LSU), KU843924 (Tef-1a).
Figure 9

Holotype. Hungary, Pest, Csévharaszt, Csévharaszti borokás, in sand steppe vegetation: *Festucetum vaginatae*, T. Knutsson, U. Andersson, T. Gunnarsson, M. Jeppson, 11 Apr. 2011, M. Jeppson 8907 (GB! paratype AH).

Etymology. The name refers to the large spores.

Description. Spore-sac subglobose, 3–8 mm. Endoperidium hyphal, encrusting sand. Endoperidium greyish white to pale greyish ochraceous with some adhering sand grains, smooth. Mouth fibrillose-fimbriate, slightly mammose (Figure 9f). Socket inconspicuous. Stem slender, 15–23 x 1–2 mm, whitish to pale yellowish brown towards the base, which is slightly widened, covered by sand and provided with a distinct pseudorhiza (Figure 9a,e). Capillitium 3–7 µm in diameter, with irregular, undulating inner walls (Figure 9b). Septa scarce, not or only very slightly widened. Spores subglobose, 5.5–7.0 µm (av. 6.5 µm), smooth under light microscopy and SEM (Figure 9c, g–i).

Habitat and distribution. Recorded in exposed sand steppe habitats in Hungary and Slovakia. Usually not very numerous in its localities.

Notes. The species reminds of the new species *T. pannonicum* in its habit, but a check under the microscope is enough to distinguish *T. grandisporum*. *Tulostoma leiosporum* R.E. Fr., described from S. America, is similar to *T. grandisporum* in macromorphology but differs in spore size (lectotype: 4.5–5.5 µm according to Moreno et al. 1997). The lectotype of *T. leiosporum* has the same type of ragged, undulating inner walls of the capillitium.

Other specimens examined. HUNGARY, **Bács-Kiskun**: Kerekegyháza, Kákás-Király tó, sandy pasture along wheel track, 16 Apr. 2009, U. Andersson, T. Gunnarsson, M. Jeppson 8924 (GB)*; Kiskunhalas, sandy grassland, 19 Jan. 2014, P. Finy 10 (GB)*; Lakitelek, Szikra, 300 m SE of railway station, along railway, on sandy path,

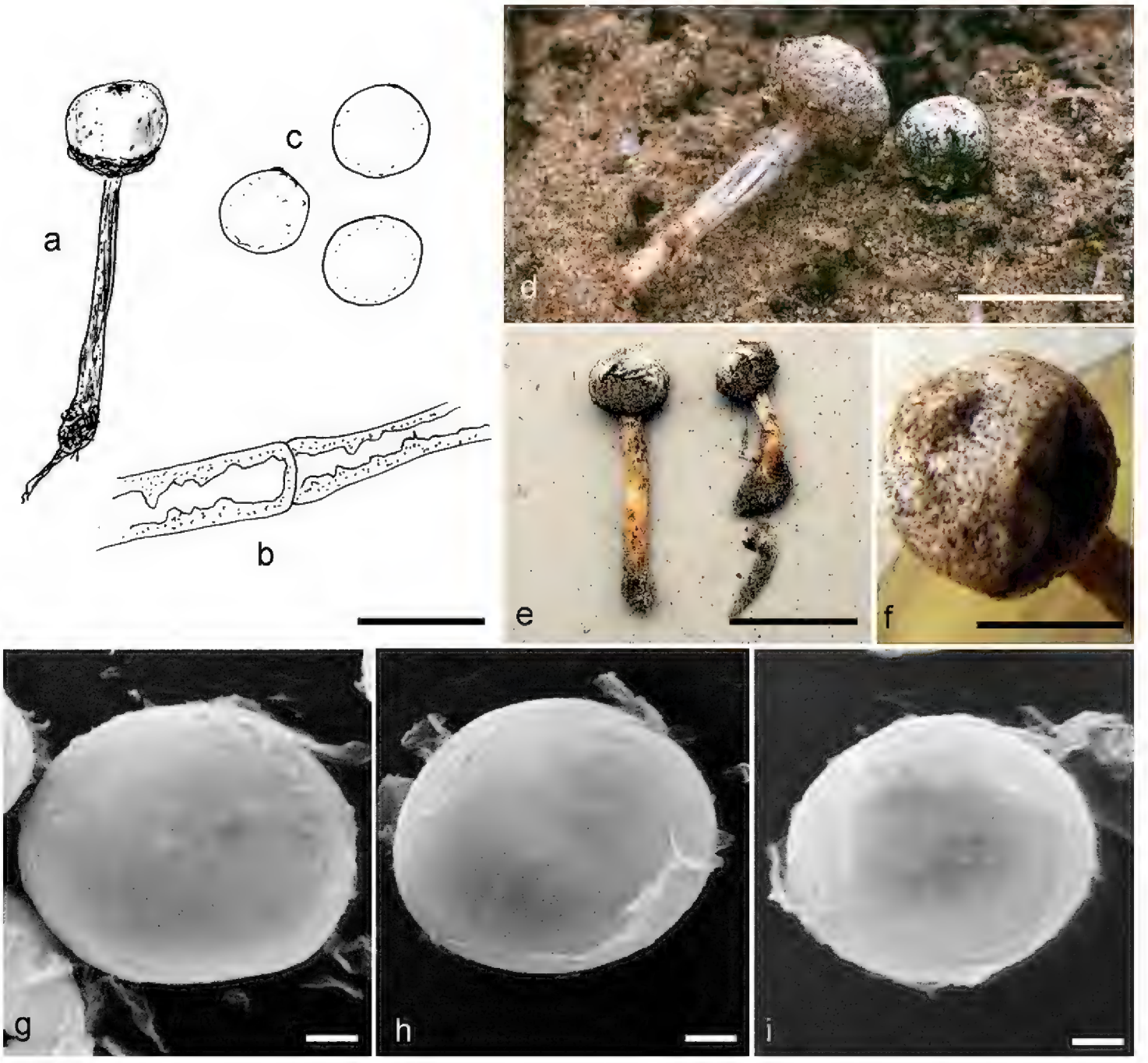

Figure 9. *T. grandisporum*, holotype (M. Jeppson 8907, GB): **a, e** basidiocarps **b** capillitium **c, g–i** spores **d** basidiocarps (M. Jeppson 7735, GB) **f** detail of spore sac showing the fibrillose-fimbriate mouth; Scale bars: **a, d–e** = 10 mm; **b–c** = 10 µm; **f** = 4 mm; **g–i** = 1 µm.

17 Feb. 2004, J. Jeppson, M. Jeppson 6633 (GB)*; Ibidem, 10 Apr. 2006, J. Jeppson, M. Jeppson 7734, 7735 (GB); **Pest**: Örkény, military training field halfway between Tatárszentgyörgy and Örkény, sand steppe vegetation, 14 Oct. 2008, T. Knutsson, E. Larsson, J. Nilsson, M. Jeppson 8793 (GB)*. SLOVAKIA, **Podunajská nížina**: Marcelová, Marcelovské piesky, on sand, 8 Dec. 2013, P. Brůžek 131208 (GB)*.

Tulostoma kotlabae **Pouzar, in Pilát, Fl. ČSR, B-1, Gasteromycetes: 598, 815. 1958.** Figures 2g, 10

Holotype. SLOVAKIA: **Záhorská nížina**: apud viam ferream haud procul loco "Krůžku", inter Kuty et Sekule, 25 Sept. 1954, F. Kotlaba (PRM 704203!)*

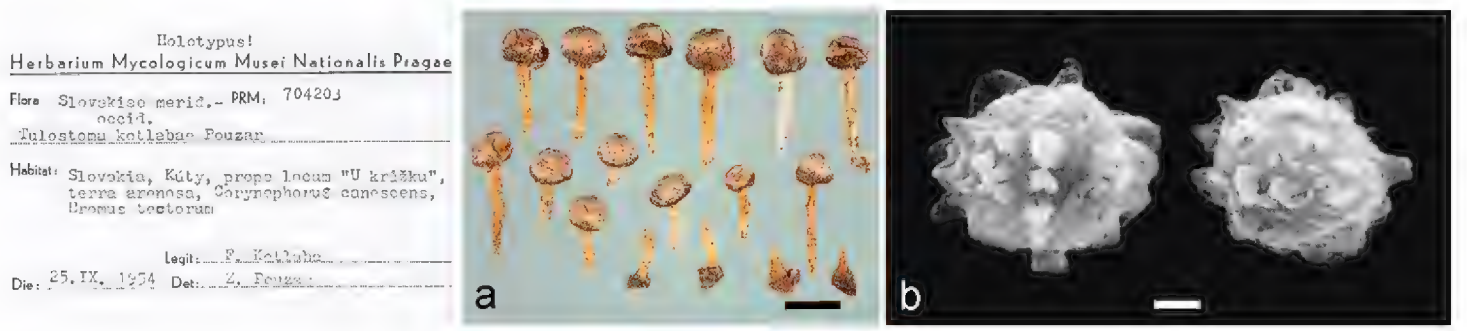

Figure 10. *T. kotlabae*, holotype (PRM 704203): **a** basidiocarps **b** spore ornamentation under SEM. Scale bars: **a** = 10 mm; **b** = 1 µm.

The studied material fits the original description as well as that of Wright (1987), and it is furthermore supported by ITS sequence data from the holotype. The eight closely related, but still unidentified, species (*T.* spp. 6-13) recovered in our phylogenetic analysis (Figure 1b) are in need of further investigation before they can be described as distinct and separate species.

Habitat and European distribution. *Tulostoma kotlabae* occurs in dry and exposed habitats such as steppe vegetation and sand dunes, preferably on calcareous, sandy soils. It is, to date, only known from Europe, from where it is on record from Denmark, France, Germany, Hungary, Lithuania, Poland, Russia (S part), Slovakia, Spain, and Sweden (Calonge 1998, Nitare 1988, Jeppson 2006a, 2008, Rimóczi et al. 2011, Rudnicka-Jezierska 1991, Winterhoff 2000, Wright 1987).

Other specimens examined. CZECH REPUBLIC, **Ústí nad Labem**: Velké Žernoseky, Kalvárie, 18 Sept. 2014, P. Brůžek 140918 (GB)*. DENMARK, **Jylland**: Hirtshals, Tversted, Vesterklit, among *Syntrichia* and *Sedum acre* on coastal sand dunes, 24 Apr. 2005, S. Jeppson, J. Jeppson, M. Jeppson 7441 (GB)*; **Møn**: Ulfshale, 2013, T. Læssøe 13035 (C). FRANCE, **Nord-France-Pas de Calais**: Étaples, Baie de la Canche, 30 Oct. 2012, E. Hultqvist, E. Larsson, M. Jeppson 9585 (GB)*. LITHUANIA, **Neringa**: Nida, 1 Oct. 1997, M. Westberg (LD). SLOVAKIA, **Kúty**: prope locum "U krůžku", terra arenosa, *Corynephorus canescens*, *Bromus tectorum*, 8 Jul. 1954, F. Kotlaba (PRM *704202*, GenBank KX576543); Plavecký Mikuláš, along road to Mikulášov, sandy road verge in pine forest, 22 Jun. 2006, M. Jeppson 7923 (GB)*. SWEDEN, **Bohuslän**: Tjärnö, Sydkoster, Kilesand, sandgräshed vid havet, 14 Apr. 2005, A.E. Torkelsen, G. Gulden, A. Karlsson, J. Nilsson, J-Å. Lönnqvist, A. Ryberg, J. Jeppson, M. Jeppson 7187 (GB)*; **Gotland**: Gammelgarn, S om Skags, sandgräshed, 26 Sept. 2001, J. Jeppson, M. Jeppson 5597 (GB)*; Gotska Sandön, gräshed med *Corynephoretum* nära stranden, 24 May 2013, T. Knutsson 2013-135 (GB); **Öland**: Högby, Dödevi, Kesnäsudden N, sandy grassland, 24 May 2007, S. Jeppson, J. Jeppson, M. Jeppson 8451 (GB); Källa, Nyby orde, 25 May 2006, M. Jeppson 7909 (GB); **Skåne**: Falsterbo, Nabben, klit med *Artemisia campestris* nær Fyrträdgården, 16 Sept. 1979, D. Boertmann 79074 (LD)*; **Skåne**: Åhus, Horna bränneri, 14 Nov. 2007, J. Jeppson, M. Jeppson 8964 (GB); Skåne, Lyngsjö, 15 Oct. 2000, S-Å. Hanson, J. Jeppson, M. Jeppson 5426 (GB)*.

Tulostoma lloydii **Bres., in Petri, Ann. Mycol. 2(5): 423. 1904.**
Figures 2h, 11

Holotype. USA (S!).

A characteristic species originally described from N. America. We have not sequenced the holotype, but its macro- and micromorphology (Figure 11a) are very similar to that of the collections here studied (Figure 2h).

Habitat and distribution. *Tulostoma lloydii* is recorded from N. America and Europe (Wright 1987). In Europe it has a Mediterranean distribution, having been reported from Italy and Spain. According to Wright (1987) it is a species of "forest soil, among humus and plant debris". The studied material from Italy was collected in a garden, but there are also findings from more xeric environments on calcareous soil and on littoral sand dunes in Spain.

Other specimens examined. ITALY, **Friuli-Venezia Giulia**: Pordenone, Vivaro, in litter under *Thuja orientalis* in church yard, Oct. 2012, E. Campo, M. Lahti 201210 (GB)*. SPAIN, **Córdoba**: Priego de Córdoba, terreno calizo con encinas, Apr. 1988, J. Gómez (AH *11606*)*; **Madrid**: Guadalix de la Sierra, entre briófitos, en suelo básico, 23 May 2003, A. González, ARBA (AH 31156, GB)*.

Tulostoma lusitanicum **Calonge & M.G. Almeida, in Calonge, Bol. Soc. Micol. Madrid 25: 302. 2000.**
Figure 12

Holotype. PORTUGAL, **Estremadura**: Pinhal do Rei, 8 Jan. 1991, G. Almeida, F. Estrella, F.D. Calonge (LISU-MGA 8!)*.

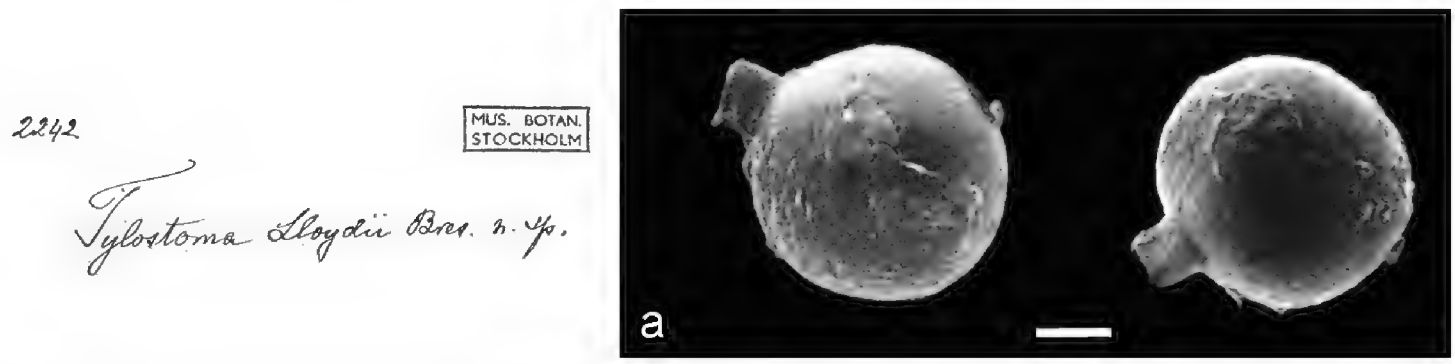

Figure 11. *T. lloydii*, holotype (S): **a** spore ornamentation under SEM. Scale bar: 1 μm.

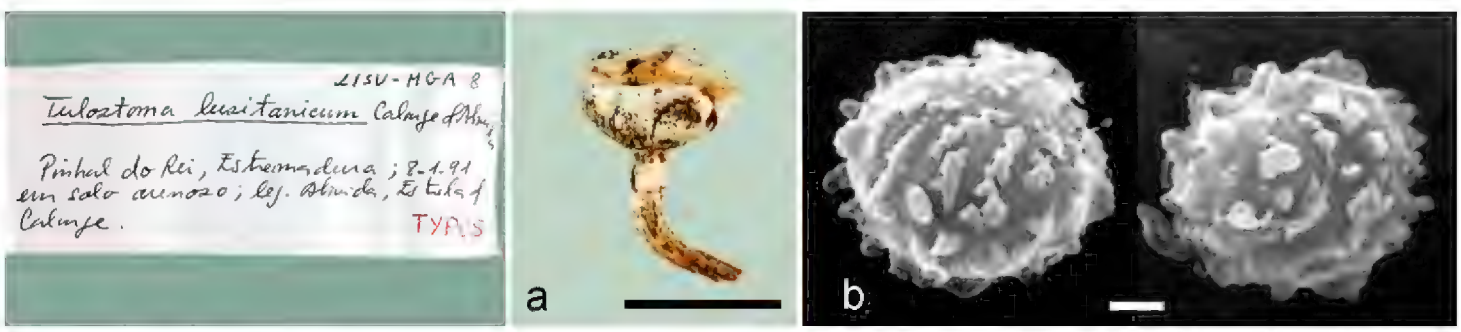

Figure 12. *T. lusitanicum*, holotype (LISU-MGA 8): **a** basidiocarp **b** spore ornamentation under SEM. Scale bars: **a** = 10 mm; **b** = 1 μm.

The species was invalidly published by Calonge and Almeida (1992) and later validated by Calonge (2000). ITS sequence data confirm it as a distinct species (Figure 1b).

Habitat and distribution. Recorded only from sand dunes in Portugal.

Tulostoma melanocyclum **Bres., in Petri, Ann. Mycol. 2(5): 415. 1904.**
Figure 2i

Holotype. ITALY: in glabrosis prope Tridentinum, 1902, G. Bresadola (K).

This species is closely related to *T. squamosum*, *T. subsquamosum,* and the new species *T. calcareum*. It is distinguished by its dark brown to almost black stipe and a prominent dark peristome contrasting to the pale colours of the surrounding endoperidium in old and weathered specimens. In young basidiomata the endoperidium is more or less orange brown and the stem ± orange brown. The spores are verrucose-echinate and normally lack anastomoses and crests as seen under SEM (Figure 2i). It can be readily distinguished from *T. brumale*, which often grows in the same locations, by its darker stem and the more coarsely ornamented spores. Photos of the holotype collection at K (habit and spore ornamentation under SEM) were included in Altés et al. (1996).

Habitat and distribution. Grows in dry sandy grasslands, on sand dunes, sand steppes and rupicolous steppe slopes. According to Wright (1987) it is a typically European species. It is, however, also recorded from Central Asia (Dörfelt and Täglich 1990). It is widespread in Europe, reaching as far north as southernmost Fennoscandia.

Specimens examined. BELGIUM, **W. Vlanderen**: Westhoek, 1 Nov. 2012, E. Hultqvist, E. Larsson, M. Jeppson 9618 (GB). CZECH REPUBLIC, **Pavlovské vrchy**: Děvin, 27 Jun. 2008, M. Jeppson 8839 (GB). DENMARK, **Jylland**: Kjul Å, 3 Aug. 1906, J. Lind (C). **Jylland**: Uggerby Å, 29 Dec. 1980, T. Læssøe (C). FRANCE, **Nord-France-Pas de Calais**: Etaples, Baie de la Canche, littoral sand dunes, 31 Oct. 2012, E. Hultqvist, E. Larsson, M. Jeppson 9596 (GB)*. HUNGARY, **Bács-Kiskun**: Kunbaracs, Csaradas, sand steppe vegetation, 2 Nov. 2002, J. Jeppson, M. Jeppson 6014 (GB); Kunbaracs, sand steppe vegetation, 1 Aug. 2008, L. Nagy, M. Jeppson 8815 (GB)*; Orgovány, Orgoványi rétek, Stipeto-festucetum on calcareous sand, 18 Apr. 2009, M. Jeppson 8974 (GB)*; **Pest**: Örkény, military training field, sand steppe vegetation, 2 Nov. 2002, J. Jeppson, M. Jeppson 6036 (GB)*. RUSSIAN FEDERATION, **Irkutskaya obl.**: Baykal Lake, Olkhon Island, Cape Unschuy, 15 Sept. 2000, H. Knudsen 00.314 (C)*. SLOVAKIA, **Nítra**: Štitáre, 29 Jun. 2009, S. Adamčik, M. Jeppson 9116 (GB); Malé Karpaty, Devinská Kobyla, Sandberg, V. Kautman, 22 Nov. 2009 (BRA); **Podunajská nížina**: Marcelova, Marcelovské Piesky, 25 Jun. 2011, I. Kautmanová, V. Kautman, M. Jeppson 9331 (GB). SWEDEN, **Öland**: Lopperstad, sandstäpp med *Carex ligerica*, 29 May 2005, A. Bohlin, K. Bohlin 050529 (GB)*; **Skåne**: Brösarp, Brösarps norra backar, 15 Dec. 2001, I. Månsson 011215 (GB)*; Degeberga, Söndre klack, 13 Mar. 2016, Ö. Fritz (GB); Maglehem, Kumlans NR, stäppartad torräng, 15 Dec. 2008, S. Hanson 2008-247 (GB)*.

Tulostoma niveum **Kers, Bot. Not. 131: 411. 1978.**
Figure 2j

Holotype. SWEDEN, **Uppland**: Djurö, Munkö, 23 Sept. 1973, L.E. Kers 4264 (S!).

Tulostoma niveum is a well-defined species with small and slender, whitish basidiomata with conically protruding mouth and distinctly membranous exoperidium. Molecular data suggest a close relationship with the new species *T. eckbladii* and *T. giovanellae* (Figure 1c).

Habitat and distribution. Occurs among mosses on ± calcareous boulders, flat rocks, or stone walls in semi-shaded localities. Recorded from Finland, Norway, Sweden, Switzerland, and the UK (Scotland). Not known from outside Europe.

Specimens examined. NORWAY, **Oslo**: Malmøykalven, among mosses on calcareous cliffs, 13 Oct. 2006, A-E. Torkelsen, A. Wollan, A. Karlsson, J. Nilsson, J. Markussen, M. Jeppson 8268 (GB). SWEDEN, **Södermanland**: Vårdinge, Usta SW, among *Tortella tortuosa* on calcareous rocks near lake shore, 13 Oct. 2005, M. Jeppson 7692 (GB)*; Utö, Notholmen, på mosstuvor på urkalkhäll nära havet, 14 Oct. 2005, I. Holst, L. Almqvist, M. Gothnier, M. Jeppson 7699 (GB)*; **Uppland**: Djurö socken, Runmarö, 300 m V om nordspetsen av ön i norra delen av Vitträsk, hällmark i norra kanten av betesmark, 1973, L.E. Kers 4775 (S); **Västmanland**: Viker, Näsmarkernas NR, Näset, on calcareous rocks among *Tortella tortuosa*, 29 Aug. 1999, J. Jeppson, M. Jeppson 5229 (GB)*. SWITZERLAND, **Genève**: Chancy, 11 Nov. 2008, J-J. Roth (GB); Ibidem, Dec. 2015 (GB).

Tulostoma obesum **Cooke & Ellis, Grevillea 6: 82. 1878.**
Figures 2k, 5c

Holotype. U.S.A., **Colorado**, Coke 2715 (K 39158); Isotype: (NY!)*

Sequenced Spanish collections were shown to be identical with the ITS sequence of the isotype from Colorada, USA (Cooke 2715, NY). With respect to morphology, *T. obesum* reminds of a species reported under *T. obesum* or *T. volvulatum* (*T. volvulatum* var. *obesum*) by Babos (1999), Calonge (1998), Hollós (1904), and Rimóczi et al. (2011) from the Iberian Peninsula and East Central Europe. However, according to molecular data, those records can be attributed to other species despite sharing a number of morphological features with *T. obesum*. SEM-photos for *T. obesum* show completely smooth spores (Figure 2k). Photos of the holotype and isotype collections at K and NY (habit and spore ornamentation under SEM) were included in Altés et al. (1999).

Habitat and distribution. A psammophilous species originally described from Colorado. In Europe it is recorded from littoral sand dunes and halophytic steppe habitats in SE Spain.

Other specimens examined. SPAIN, **Almería**: Cabo de Gata, La Salina, NW part, among halophilous vegetation on sand, 21 Apr. 2008, M. Jeppson 8695 (GB)*; **Almería**: Tabernas, Puente de los Callejones, among halophilous vegetation on sandy

rambla, 16 Feb. 2003, J. Jeppson, M. Jeppson 6305 (GB); Ibidem, 22 Apr. 2008, M. Jeppson 8707 (GB)*; Ibidem, 22 Apr. 2008, M. Jeppson 8708 (GB); **Castellón**: Oropesa del Mar, dunas litorales con gramíneas, 8 Dec. 1995, J. Ayllón (AH 20901, GB)*.

Tulostoma pannonicum **Jeppson, Altés, G. Moreno & E. Larss., sp. nov**.
MycoBank: MB819403
GenBank: KU519010 (ITS-LSU)
Figure 13

Holotype. Hungary, Bács-Kiskun, Fülöpháza, Fülöpházi Homokbuckák, N of road 52, sand steppe vegetation, Leg: J. Jeppson, M. Jeppson, 11 Apr. 2006, M. Jeppson 7764 (GB!, isotype AH).

Etymology. The name refers to Pannonia, an ancient Roman province in Central Europe, where this species was first collected.

Description. Spore-sac subglobose, 4–7 mm in diameter. Exoperidium indistinct to thinly membranous, incrusting sand, remaining as a cup-like structure at the base of the spore-sac (Figure 13d). Endoperidium greyish white, smooth, or pitted from detached sand grains. Mouth indistinct to fibrillose-fimbriate (Figure 13f). Socket inconspicuous, slightly separated. Stem slender, 10–20 x 1–2 mm, ochraceous to pale greyish, smooth, somewhat longitudinally furrowed, with a slightly widened base and a distinct pseudorhiza (Figure 13a,d). Mature gleba ferrugineous. Capilltium 3–5 µm in diameter with even inner walls and abundant, strongly widened septa (Figure 13b). The capillitium easily breaks up at the septa. Spores subglobose to broadly ellipsoid, 3.5–4.5 µm (av. 4.0 µm), smooth. Under SEM the spore wall is smooth to slightly rugulose (Figure 13g-i).

Habitat and distribution. The species is recorded from exposed sand steppe habitats on calcareous soil in Hungary.

Notes. Rimóczi et al. (2011) recorded this species from Hungary as *T. leiosporum* and noted that the spores were slightly smaller than those of the lectotype of *T. leiosporum,* deposited in Herbarium S. Some photos of the lectotype collection of *T. leiosporum* (habit and spore ornamentation under SEM) were included in Moreno et al. (1997).

Other specimens examined. HUNGARY, **Bács-Kiskun**: Fülöpháza, Fülöpházi Homokbuckák, S of road 52, sand steppe vegetation at forest edge (*Pinus*), 12 Oct. 2008, E. Larsson, J. Nilsson, T. Knutsson, M. Jeppson 8715 (GB)*; Orgovány, Orgományi rétek, sand steppe vegetation, 17 Jun. 1999, M. Jeppson 990617a (GB)*; Ibidem, among *Syntrichia* on sand, 12 Apr. 2006, J. Jeppson, M. Jeppson 7803 (GB)*.

Tulostoma pseudopulchellum **G. Moreno, Altés & J.E. Wright, Mycotaxon 43: 481. 1992.**

Holotype. SPAIN, **Madrid**: Alcalá de Henares, junto al cerro Ecce Homo, vegetación esteparia con yesos, Mar. 1989, C. Illana, G. Moreno, A. Altés (AH 11605!)*.

Figure 13. *T. pannonicum*, holotype (M. Jeppson 7764, GB): **a, d–e** basidiocarps **b** capillitium **c, g–i** spores **f** detail of spore sac showing the fibrillose-fimbriate mouth (M. Jeppson 7803, GB). Scale bars: **a, d–e** = 10 mm; **b–c** = 10 µm; **f** = 5 mm; **g–i** = 1 µm.

Tulostoma pseudopulchellum is a small and slender species with a macro-morphological resemblance to *T. pulchellum*. It differs from that species by having significantly smaller basidiomata and irregularly rugulose spores (SEM). It has a distinctly membranous exoperidium and a fibrillose-fimbriate mouth. Moreno et al. (1992b) suggested a close relationship with *T. pulchellum* but this could not be confirmed in the molecular analyses.

Habitat and distribution. So far only known from its type locality in dry gypsum slopes at Alcalá de Henares in Central Spain.

Other specimens examined. SPAIN, **Madrid**: Alcalá de Henares, junto al cerro Ecce Homo, vegetación esteparia con yesos, Mar. 1989, C. Illana, G. Moreno, A. Altés(paratype, AH 11603)*; Ibidem, junto al cerro Malvecino, en margas yesíferas, 3 Nov. 1989, *G.* Moreno, J.L. Manjón, A. Altés (AH 11699)*.

***Tulostoma pulchellum* Sacc., Bull. Soc. Mycol. France 5(4): 118. 1890 [1889].**
Figures 2l, 14, 15

≡ *Tulostoma poculatum* V.S. White, Bull. Torrey Bot. Club 28: 431. 1901. Holotype. USA, Nebraska: Long Pine, Bates (NY).
≡ *Tulostoma minutum* V.S. White, Bull. Torrey Bot. Club 28: 433. 1901. Holotype. USA, Colorado: Bethel 22a (NY).
≡ *Tulostoma hollosii* Z. Moravec, Preslia 28: 36. 1956. Holotype. CZECH REPUBLIC, Praha: Prokopské údolí, kopec u koupalisté, 3 Oct. 1952, Moravec (PRM).

Holotype. AUSTRALIA, Tepper (PAD).

This is a characteristic species with a distinct membranous exoperidium, a snow-white endoperidium and a ± delimited, fibrillose-fimbriate mouth. The ornamentation of the spores is composed of a dense cover of low verrucae, sometimes coalescing into ridges. Collections with a ridged type of ornamentation have been distinguished as *T. hollosii* and *T. pulchellum* var. *subfuscum* (cfr. Moreno et al. 1992b, Specht and Schubert 2012). Our sequence data based on specimens with isolated verrucae as well as on those with distinct ridges (Figure 2l), a paratype of *T. hollosii* included (Figure 15), indicate conspecificity. Our molecular data suggest *T. pulchellum* to be closely related to *T. striatum* (Figure 1b).

Habitat and distribution. *Tulostoma pulchellum* has a cosmopolitan distribution, occurring in exposed, dry, and warm habitats. In Europe it is on record from the Czech Republic, Germany, Hungary, Italy, Romania, Slovakia, and Spain (Kreisel 2001, Kříž 2011, Specht and Schubert 2012, Sarasini 2005). It is here confirmed also from Central Asia (Mongolia).

Specimens examined. CZECH REPUBLIC, **Litomeřice**: Radobýl, 18 Jan. 2014, P. Brůžek (GB); **Praha**: in valle Prokopské údoli dicta, ap. vicum Klukovice prope Pragam, in sinuis saxorum calcarearum, 2 Dec. 1951, Z. Moravec 132/51 (paratype *T. hollosii*, UPS)*. GERMANY, **Sachsen-Anhalt**: Halberstadt, Hammelberg, Nov. 2011, H. Schubert, P. Specht (GB). HUNGARY, **Bács-Kiskun**: Bugac, Bugacpuszta, near Pásztormuzeum, *Cynodon*-grassland, 12 Apr. 2006, I. Rimóczi, T. Knutsson, T. Gunnarsson, J. Jeppson, M. Jeppson 7788 (GB); Kecskemét, Kisfái, sandstäpp i kanten av tallplantering, 4 Feb 2004, J. Jeppson, M. Jeppson 6589 (GB); Kunbaracs, sand steppe vegetation, 10 Oct. 2004, K. Hjelmér, M. Jeppson 6812 (GB); Lakitelek, Szikra, by the railway station, ruderal sand steppe vegetation, 17 Feb. 2004, J. Jeppson, M. Jeppson 6647 (GB)*; Lakitelek, Árpádszállás, sandy roadside in *Pinus* plantation, 17 Feb. 2004, J. Jeppson, M. Jeppson 6614 (GB); **Csongrád**: Opusztaszer, Hantháza, ruderal, sheep-grazed sand steppe vegetation, 14 Apr. 2006, J. Jeppson, M. Jeppson 7833 (GB)*; **Pest**: Csévharaszt, Csévharaszti borókás, *Festucetum vaginatae,* 11 Apr. 2009, T. Knutsson, U. Andersson, T. Gunnarsson, M. Jeppson 8905 (GB). MEXICO, **Baja California**: La Rumorosa, terreno árido, escarpado y pedregoso, 9 Dec. 1987, C. Ochoa, A. Altés (AH 12967, GB)*. MONGOLIA, Terelj National Park, 2010, Ö. Fritz 2010-1 (GB)*. SPAIN, **Granada**: Cenes de la Vega, cerca del Llano de la Perdiz,

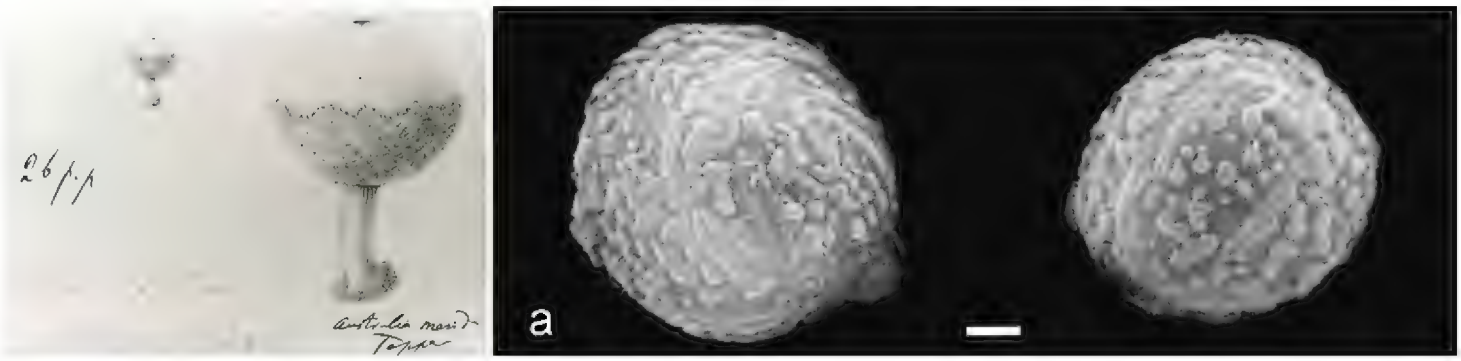

Figure 14. *T. pulchellum*, holotype (PAD): a. spore ornamentation under SEM. Scale bar: 1 µm.

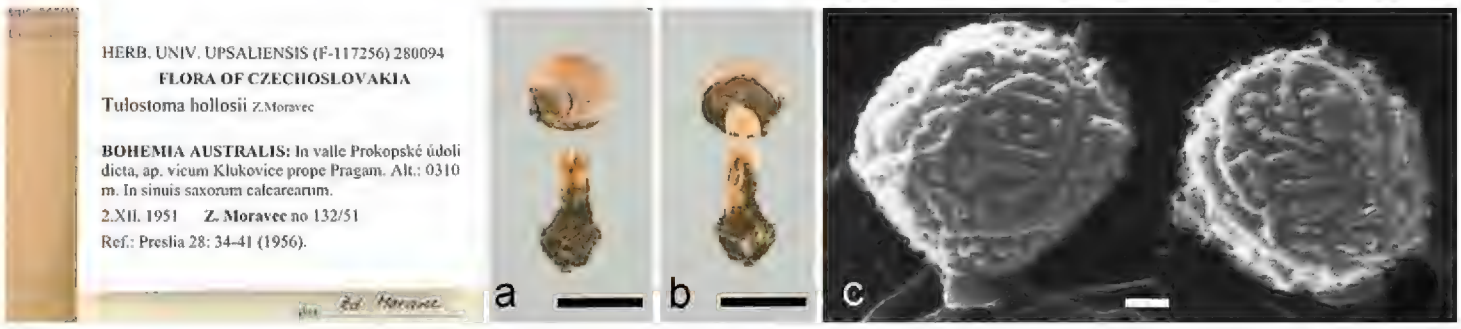

Figure 15. *T. hollosii*, paratype (Moravec 132/51, UPS): **a–b** basidiocarp (single, both sides) **c** spore ornamentation under SEM. Scale bars: **a–b** = 10 mm; **c** = 1 µm.

zona árida con *Pinus halepensis*, *P. sylvestris* y *Cistus ladanifer*, 21 Jul. 1991, R. Galán (as *T. pulchellum* var. *subfuscum*, AH *13627*, GenBank KX640982); **Segovia**: Cuellar, en suelo arenoso en claro de pinar de *Pinus pinaster*, 20 Nov. 1994, F. García (MA-F *33568*)*; Valladolid, Tudela de Duero, Pradillos, suelo arenoso bajo *Pinus pinea*, 27 Feb. 1993, P. Juste (MA-F *32338*, GenBank KX640983).

Tulostoma punctatum **Peck, Bull. Torrey Bot. Club 23(10): 419. 1896.**
Figures 5d, 16a, 17, 18

≡ *Tulostoma fimbriatum* var. *punctatum* (Peck) J.E. Wright, Biblioth. Mycol. 113: 115. 1987.
≡ *Tulostoma subfuscum* V.S. White, Bull. Torrey Bot. Club 28: 433. 1901. Holotype. USA, Colorado: Denver, 16 Feb. 1896, Bethel 21 (NY!)*;

Lectotype. USA, Kansas: Rooks Co., 23 Jul. 1896, E. Bartholomew (BPI 729033!)*.

The first European records of this traditionally North American species are here confirmed. The species can easily be confused with *T. fimbriatum*, but is distinguished by the somewhat smaller spores that are more ornamented with a verrucose-spiny ornamentation without ridges (Figure 16a). Study of the macro-micromorphology and sequence data of the type collections of *T. punctatum* (Figure 17) and *T. subfuscum* (Figure 18), indicate conspecificity. It is surprising to see that the spore ornamentation of the type of *T. subfuscum* is very different from the expected one (cfr. description and illustrations by Wright 1987). The presence of distinctive punctate pits on the

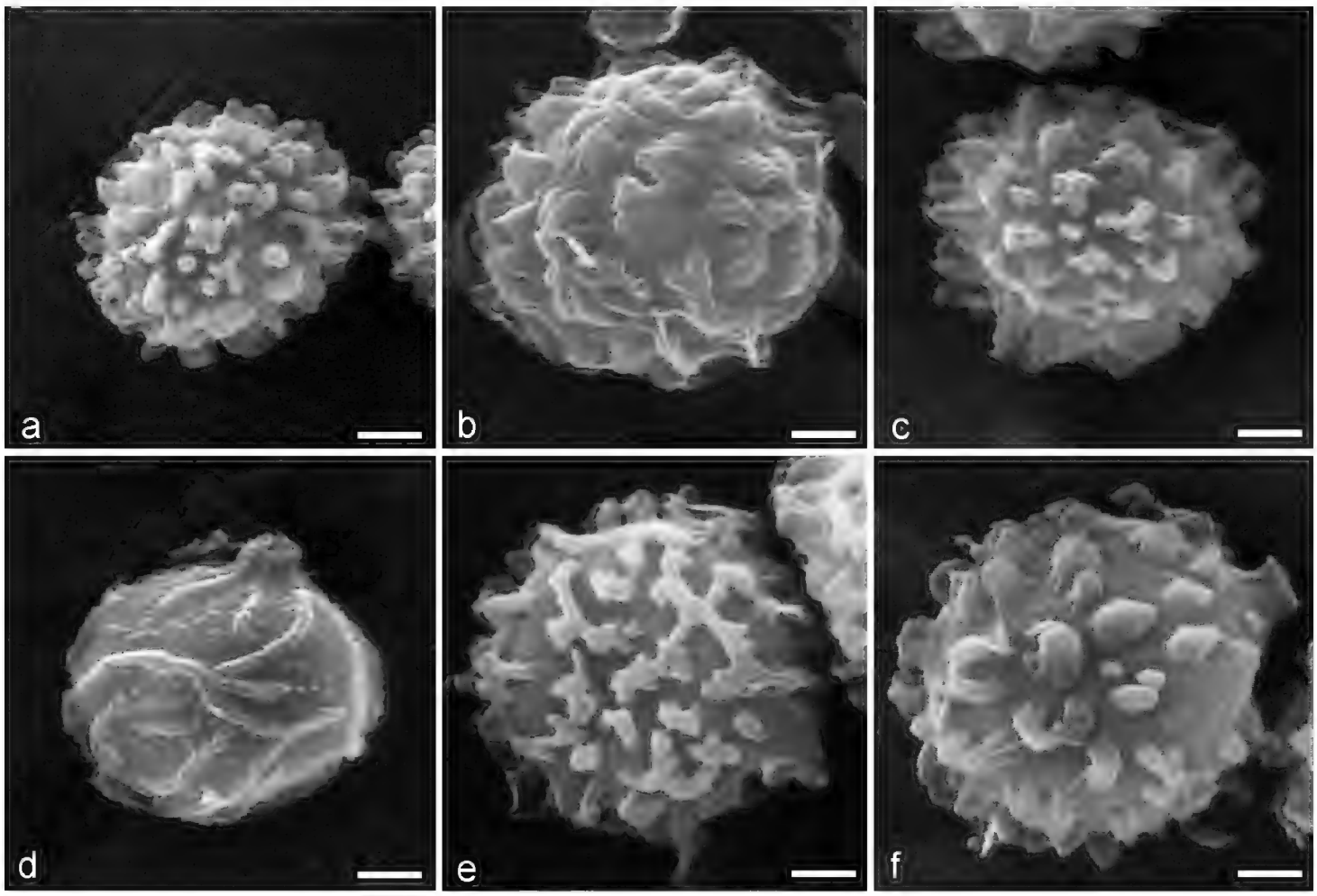

Figure 16. Spore ornamentation under SEM of European species (cont.): **a** *T. punctatum* (M. Jeppson 7472, GB) **b** *T. simulans* (M. Jeppson 9302, GB) **c** *T. squamosum* (AH 15483) **d** *T. striatum* (AH 15543) **e** *T. subsquamosum* (M. Jeppson 3475, GB) **f** *T. winterhoffii* (M. Jeppson 2379, GB). Scale bars: 1 µm.

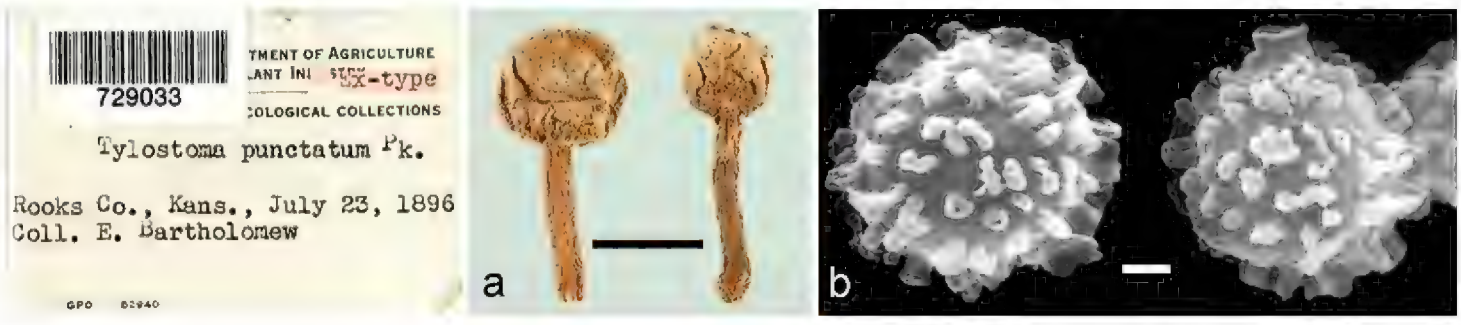

Figure 17. *T. punctatum*, lectotype (BPI 729033): **a** basidiocarps **b** spore ornamentation under SEM. Scale bars: **a** = 10 mm; **b** = 1 µm.

endoperidium of *T. punctatum*, as described by Peck (1896), could not be observed in the European specimens. A somewhat pitted appearance of the endoperidium is a common, but inconsistent, feature among species of *Tulostoma*, and of little taxonomic value. The pits are scars or depressions caused by sand grains adhering to the peridium in early stages of development.

Habitat and distribution. Originally described from the USA (Kansas). In Europe on sandy soil on exposed, anthropogeneous sites (e.g. church lawns/car parks, abandoned orchards) in SW Slovakia.

Other specimens examined. SLOVAKIA, **Záhorská nížina**: Vel'ke Leváre, by the church, grassy road side on sandy soil, 2 Oct. 2005, J. Jeppson, M. Jeppson 7472

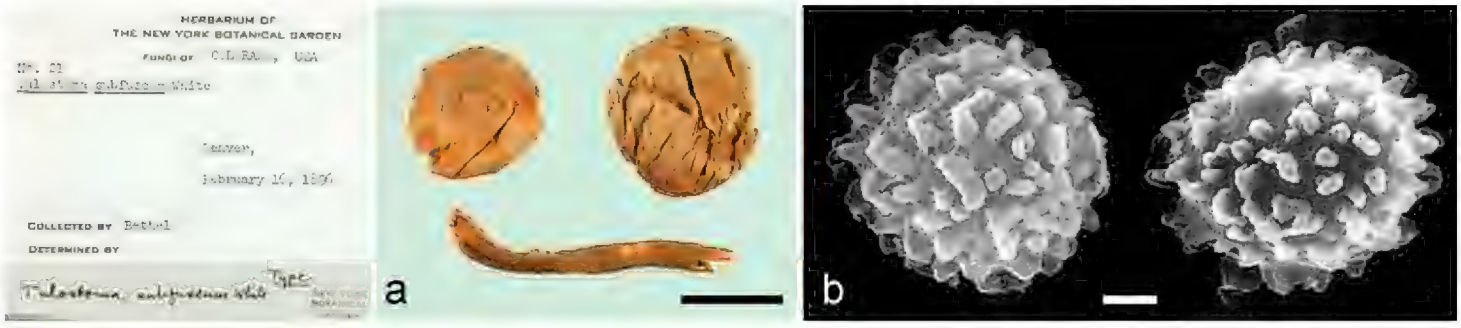

Figure 18. *T. subfuscum*, holotype (Bethel 21, NY): a. basidiocarps **b** spore ornamentation under SEM. Scale bars: a = 10 mm; b = 1 µm.

(GB)*; Stupava, in dry sandy grassland in an abandoned orchard, 16 Oct. 2014, T. Knutsson, E. Larsson, R. Bednar, V. Kautman, I. Kautmanová, A. Košuthová, M. Jeppson 10058 (GB)*.

Tulostoma simulans **Lloyd, The Tylostomeae: 18. 1906.**
Figures 5e–f, 16b, 19

≡ *Tulostoma moravecii* Pouzar, in Pilát, Fl. ČSR, B-1, Gasteromycetes: 597, 814. 1958. Holotype. CZECH REPUBLIC, Praha: inter Homolka et Lochkov, 21 Sept. 1952, Z. Moravec (PRM).

Holotype. USA, **Texas**: Denton, Long (herb. Lloyd 13636, BPI 704611).

As was indicated already by its author (Lloyd 1906), this species "simulates" other species of *Tulostoma*, particularly *T. brumale*, from which it differs by having a hyphal exoperidium and larger spores. According to our molecular results it is known from Austria, Hungary, Spain, Sweden, and the UK (a British sequence was kindly provided by Martyn Ainsworth, Kew, but is not shown in the tree). Sequence data of the European specimens, labelled as *T. beccarianum*, *T. brumale*, and *T. moravecii* (isotype, Figure 19), cluster with a paratype in PC of *T. simulans*. Unfortunately the holotype material of *T. simulans* was not available for DNA extraction.

SEM-photos of the collections used in our study show irregular, rather dense, broadly based conical verrucae (Figure 16b), thus closely coinciding with the holotype material of *T. simulans* (cfr. Moreno and Altés 1992). Altés and Moreno (1993) synonymized *T. simulans* with *T. beccarianum* based on morphological similarities, but sequence data show that they should be regarded as separate species.

Habitat and distribution. N. America, S. America, Australia, New Zealand, Europe, and Asia (Kazachstan). It is here confirmed also from eastern Russia (Altay Republic). It has been found in a wide range of habitats ("forest soil, among litter, sometimes dunicolous, tree nurseries" according to Wright 1987).

European occurrences are from sand steppe habitats, sand dunes, gypsum hills, rupicolous steppe meadows, and urban plantations.

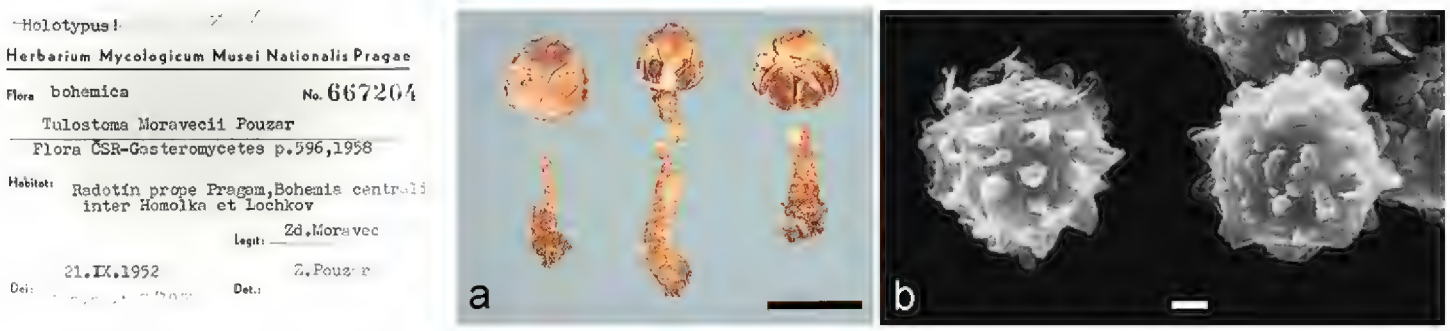

Figure 19. *T. moravecii*, isotype (PRM 667204): **a** basidiocarps **b** spore ornamentation under SEM. Scale bars: **a** = 10 mm; **b** = 1 μm.

Specimens examined. AUSTRIA, **Burgenland**: Brietenbrunn, Thenau NSG, 24 Oct. 1989, E. Mrazek 1313 (Herb. Mrazek, GB)*; Ibidem, dry calcareous grassland, 9 Oct. 2005, J. Jeppson, M. Jeppson 7865 (GB)*. CZECH REPUBLIC, **Pardubice**: Svitavy, under *Picea pungens*, 14 Feb. 2014, P. Brůžek 140214 (GB)*; **Praha-Dejvice**: Zahrada Hanspaulka, sandy soil, 1 Dec. 2013, M. Brtník, P. Brůžek 131201 (GB)*; **Praha**: inter Homolka et Lochkov, 21 Sept. 1952, Z. Moravec (isotype *T. moravecii*, PRM 667204)*. HUNGARY, **Bács-Kiskun**: Fülöpháza, Fülöpházi Homokbuckák, sand steppe vegetation, 13 Jun. 1999, J. Jeppson, M. Jeppson 4902 (GB)*; Ibidem, 17 Jun. 1997, S. Jeppson, J. Jeppson, M. Jeppson 3844 (GB)*; Ibidem, 26 Jun. 2001, M. Vetter, M. Jeppson 5497 (GB)*; **Pest**: Tatárszentgyörgy, Sarlósárpuszta, in sand steppe vegetation, 21 Feb. 2004, J. Jeppson, M. Jeppson 040221 (GB)*. RUSSIAN FEDERATION, **Altay Rep.**: Chuya River/Kuektanar River, 31 km W of Kosh-Agach on M52, 21 Aug. 2001, H. Knudsen 01.70m (C)*. SPAIN, **Madrid**: Alcalá de Henares, cerros de Alcalá, gypsophilous steppe vegetation, on road verge, 2 Mar. 2011, G. Moreno, A. Altés, A. Stridvall, E. Larsson, M. Jeppson 9302 (GB)*; Alcalá de Henares, Campus Universidad Alcalá, en terreno básico (arcilloso), junto a camino, vegetación ruderal, 10 Mar. 1993, V. González (AH 15633, GB)*; **Zaragoza**: Bujaraloz, Laguna de la Muerte, in halophilous steppe vegetation at lake shore, 5 Apr. 2010, E. Larsson, U. Andersson, T. Gunnarsson, T. Knutsson, M. Jeppson 9064 (GB)*. SWEDEN, **Skåne**: Brösarp, Torparebron, 12 Oct. 1996, M. Jeppson 3871 (GB)*. USA, **Ohio**: Granville, W.W. Stockberger (paratype *T. mammosum* var. *simulans*, PC)*.

***Tulostoma squamosum* (J.F. Gmel.) Pers., Syn. Meth. Fung. (Göttingen) 1: 139. 1801.** Figure 16c

≡ *Lycoperdon squamosum* J.F. Gmel., Systema Naturae, Edn 13 2(2): 1463 (1792). Holotype. ITALY, cl. Targioni (herb. Persoon, L).
≡ *Tulostoma verrucosum* Morgan, J. Cincinnati Soc. Nat. Hist. 12(4): 164. 1890. Lectotype. USA, Ohio: Cincinatti, 1883, A.P. Morgan (BPI).
≡ *Tulostoma mussooriense* Henn., Hedwigia 40: 337. 1901. Holotype. INDIA, Mussoorie: Arnigadh, 5500 ft alt., 9 Aug. 1900, W. Gollan (S!).

Remarks. *Tulostoma squamosum* is morphologically well characterized by its brown, rather tall and squarrose stem combined with a brownish warty exoperidium that upon

dehiscense leaves a distinct reticulate pattern. The exoperidium is composed of brownish sphaerocysts. The spore ornamentation under SEM (Figure 16c) shows spines fused at their tops into small pyramidal groups, sometimes coalescing to form short crests. In Scandinavia the name *T. squamosum* has previously been associated with the proposed new species *T. calcareum*, described above (cfr. ArtDatabanken 2015, Nitare 1997),

Habitat and distribution. Usually found among herbs in semi-shaded places, including open deciduous forests and gardens. Taking into account its recently established synonyms *T. mussooriense* and *T. verrucosum* (Moreno et al. 1992a), it has a cosmopolitan distribution. It seems to be one of the more widespread *Tulostoma* species in Europe, reaching north to Germany (Kreisel 1984).

Specimens examined. AUSTRIA, **Niederösterreich**: Schönfelder (bewaldete) Düne, 12 Sept. 1989, E. Mrazek 1300 (Herb. Mrazek, GB)*. FRANCE, **Nord-France-Pas de Calais**: Etaples, Garennes, Reserve nationale de la Canche, 11 Nov. 2006, E. Larsson 260-06 (GB)*. SLOVAKIA, **Malé Karpaty**: Bratislava, Devinska Kobyla, 13 Nov. 1997, Ortová 971113 (BRA, GB)*. SPAIN, **Castellón**: cerca de Morella, on calcareous soil under *Pinus nigra* and *Quercus rotundifolia*, 21 Oct. 1992, M. Honrubia (AH 15483, GB)*.

***Tulostoma striatum* G. Cunn., Proc. Linn. Soc. New South Wales 50(3): 255. 1925.**
Figure 16d

Holotype. AUSTRALIA, N.S.W: Forbes, 13 Aug. 1915, J.B. Cleland (ADW *5867*).

As was noted by Cunningham (1925), this species (originally described from Australia) strongly reminds of *T. pulchellum* (as *T. poculatum*), but can be readily distinguished by its striate spores (Figure 16d). The molecular results confirm it is a distinct species (Figure 1b).

Habitat and distribution. It is widespread in the Americas, South Africa and Australia (Wright 1987). Altés and Moreno (1991) recorded it from Europe (Spain), where it to date is only known from the city of Madrid. Asai (2004) reported it from Japan and it is now also confirmed to occur in Mongolia. According to Wright (1987) and Calonge (1998) it is a psammophilous species.

Specimens examined. MONGOLIA, Terelj National Park, 2010, Ö. Fritz 2010-2 (GB)*. SPAIN, **Madrid**: Carabanchel Alto, en arena de camino, junto a una pradera, 19 Jan. 1993, V. González (AH 15543, GB)*; Ibidem, 8 Nov. 1989, A. Martínez (AH 11991, GenBank KY000097).

***Tulostoma subsquamosum* Long & S. Ahmad, Farlowia 3: 241. 1947.**
Figures 5g, 16e

Holotype. INDIA, Gurdaspur: Jaggatpur, Aug. 1938, S. Ahmad 258 (BPI).

Tulostoma subsquamosum is characterized by a hyphal exoperidium with scattered sphaerocyst-like cells. The spores are verrucose-echinate with a tendency to form a

subreticulum (Figure 16e). It seems to be one of the more common species in the sand steppe vegetation of Central Hungary, from where it was traditionally recorded as *T. squamosum* (cfr. Hollós 1904, Rimóczi et al. 2011). In the molecular analyses it comes out as a sister species to *T. squamosum* (Figure 1c).

Habitat and distribution. Occurs in dry grasslands on sandy-clayish soil. It has a wide distribution in Asia (India, Pakistan), N and S America and Europe (Hungary, Slovakia, Spain).

Specimens examined. HUNGARY, **Bács-Kiskun**: Fülöpháza, Fülöpházi Homokbuckák, 1 Nov. 2002, J. Jeppson, M. Jeppson 6002 (GB)*; Jakabszállász, Alsójáras, 15 Feb. 2004, J. Jeppson, M. Jeppson 6563 (GB)*; Orgovány, Hattarcsarda, sand steppe vegetation, 24 Sept. 1995, A. Bohlin, K. Bohlin, M. Jeppson 3475 (GB); Orgovány, Orgoványi rétek, sand steppe vegetation, 16 Jun. 1999, S. Jeppson, J. Jeppson, M. Jeppson 4956 (GB)*; Ibidem, 15 Jun. 1999, S. Jeppson, J. Jeppson, M. Jeppson 4945 (GB)*. SLOVAKIA, **Podunjaská nížina**: Imeľ, Liscie Diery NR, sand steppe vegetation, 26 Jun. 2011, I. Kautmanová, V. Kautman, M. Jeppson 9336 (GB)*. SPAIN, **Madrid**: Ciempozuelos, yesiferous steppe vegetation, 22 Nov. 1989, J. Jeppson, M. Jeppson (GB); **Toledo**: Ocaña, in *Pinus halepensis* plantation on sandy soil, 4 Mar. 2011, G. Moreno, E. Larsson, A. Stridvall, M. Jeppson 9305 (GB)*; **Zaragoza**: Bujaraloz, terreno básico con yeso, bajo *Juniperus thurifera*, 31 Oct. 1995, G. Moreno, A. Altés (AH 19024, GB)*.

***Tulostoma winterhoffii* H. Schub. & P. Specht, in Specht and Schubert, Z. Mykol. 79(2): 421. 2015.**
Figures 5h, 16f, 20

≡ *Tulostoma fimbriatum* var. *heterosporum* J.E. Wright, Biblioth. Mycol. 113: 113. 1987. Holotype. THE NETHERLANDS, Wassenaar:"Voorlinden", 7 Oct. 1950, R.A. Maas Geesteranus 7513 (L!)*.

Holotype. GERMANY, Rheinland-Pfalz: Hohfels near Grünstadt, 24 May 1987, W. Winterhoff (HAL).

Tulostoma winterhoffii was recently described from Germany. The holotype was not available for sequencing in this study but a paratype, provided by its authors, was sequenced and found to match sequence data of the type material of *T. fimbriatum* var. *heterosporum*. In macro-morphology (Figure 5h) *T. winterhoffii* strongly reminds of *T. fimbriatum* but important features to distinguish it are the verrucose-echinate spores (no anastomoses seen under SEM; Figure 16f) and the variation in spore size (Wright 1987: up to 9.7 µm in diameter; Specht and Schubert (2013): (4.5–) 5.5–8.5 (–10.3) µm including ornamentation). Our own material shows a similar variation, averaging 5.7 – 6.1 µm, some spores reaching 9 µm. The same can be observed in the type collection of *T. fimbriatum* var. *heterosporum* (Figure 20). Based on morphology

Figure 20. *T. fimbriatum* var. *heterosporum*, holotype (L 64172): **a** basidiocarps **b** spore ornamentation under SEM; spore size variation. Scale bars: **a** = 10 mm; **b** = 1 µm.

and sequence data, *T. fimbriatum* var. *heterosporum* must thus be considered a distinct species, for which the name *T. winterhoffi* has priority.

Habitat and distribution. Mostly in dry and sandy habitats in exposed places, on more or less calcareous soils. Currently known from the Czech Republic, Denmark, France, Germany, Hungary, the Netherlands, Norway, Spain, and Sweden. In Scandinavia it reaches 60 ' N. The world distribution comprises Europe, Asia (Caucasus), and N. America (California) according to Wright (1987; as *T. fimbriatum* var. *heterosporum*).

Specimens examined. DENMARK, **Sjælland**: Greve, Hundige Strand, 1 Jan. 2015, S.O. Jensen, Atlasprojekt SOJ2105-699039 (C)*. BULGARIA, **Nesebar**: Slyanchev Brjag, 18 Sept. 1979, S. Jeppson. M. Jeppson 1359 (GB). GERMANY, **Rheinland-Pfalz**: Grünstadt, Asselheim, am Höllenberg, 12 Apr. 2013, ZFM79(2) (paratype, H. Schubert and P. Specht private herbarium)*. HUNGARY, **Bács-Kiskun**: Fülöpháza, Fülöpházai Homokbuckák, among *Syntrichia* in sand steppe vegetation, 11 Apr. 2006, L. Nagy, T. Gunnarsson, T. Knutsson, J. Jeppson, M. Jeppson 7761 (GB)*. NORWAY, **Buskerud**: Hurum, Nærnes, Grundviksveien, 3 May 2010, A-E. Torkelsen, G. Gulden, A. Karlsson, J. Nilsson, M. Jeppson 9537 (GB); **Oslo**: Græsholmen, 12 May 2006, A-E. Torkelsen, A. Karlsson, J. Nilsson, A.K. Wollan, A. Ryberg, M. Jeppson 7895 (GB, O); Munkedammen, N.M. Blytt 229 (O F-*86871*). SPAIN, **Madrid**: en césped, Dec. 1984, A.M. Moscardó (AH *11647*, GenBank KC333060); Madrid, en un solar urbano, 1988, F.J. Martínez (AH *11656*, GenBank KC333061). SWEDEN, **Bohuslän**: Tjärnö, Syd-Koster, Kilesand, sandgräshed med skalrik sand nära havet, 18 Oct. 1987, S. Jeppson, J. Jeppson, M. Jeppson 2379 (GB)*; **Gotland**: Ardre, Kungens sal, 22 Sept. 2000, J-O. Tedebrand (UPS).

New species identified in this study that lack scientific names

***Tulostoma* sp. 1**: HUNGARY, **Bács-Kiskun**: Fülöpháza, Fülöpházai Homokbuckák, sand steppe vegetation, 11 Apr. 2006, J. Jeppson, M. Jeppson 7762 (GB)*.

***Tulostoma* sp. 2**: SPAIN, **Almería**: Tabernas, Puente de los Callejones, halophilous vegetation in rambla, 22 Apr. 2008, M. Jeppson 8701, (GB)*. Ibidem, 25 Apr. 2008, J. Vesterholt, M. Jeppson 8710 (GB)*.

***Tulostoma* sp. 3**: HUNGARY, **Bács-Kiskun**: Bugac, Ösborokás, 15 Jun. 1999, S. Jeppson, J. Jeppson, M. Jeppson 4935 (GB)*.

***Tulostoma* sp. 4**: HUNGARY, **Budapest**: Bikás, sandy grassland, 2 Nov. 2011, P. Finy 12 (GB)*.

***Tulostoma* sp. 5**: SLOVAKIA, **Záhorská nížina**: Malacky, Kozánka, sandy soil, 7 Dec. 2013, I. Tomaševská, P. Brůžek 131207 (GB)*. Ibidem, 16 Oct. 2014, E. Larsson, R. Bednar, V. Kautman, M. Jeppson 10060 (GB)*.

***Tulostoma* sp. 6**: HUNGARY, **Bács-Kiskun**: Fülöpháza, Fülöpházi Homokbuckák, sand steppe vegetation, 31 Oct. 2002, J. Jeppson, M. Jeppson 5996 (GB)*.

***Tulostoma* sp. 7**: HUNGARY, **Bács-Kiskun**: Nyárlörincs, W. of Szikra, 17 Feb. 2004, J. Jeppson, M. Jeppson 6623 (GB)*. Pest, Tatárszentgyörgy, sand steppe, 11 Nov. 2014, P. Finy 1 (GB)*.

***Tulostoma* sp. 8**: HUNGARY, **Bács-Kiskun**: Bugac, Ösborokás, sandstäpp, i barrmatta under *Juniperus*, 19 Jun. 1997, S. Jeppson, J. Jeppson, M. Jeppson 3830 (GB)*. Ibidem, 12 Apr. 2006, I. Rimóczi, L. Benedek, T. Knutsson, T. Gunnasson, J. Jeppson, M. Jeppson 7795, (GB)*; Kerekegyháza, Borovicka dülö, ruderal sand steppe, roadside, 4 Nov 2002, J. Jeppson, M. Jeppson 6081 (GB)*.

***Tulostoma* sp. 9**: HUNGARY, **Bács-Kiskun**: Fülöpháza, Fülöpházi Homokbuckák, along road 52 at km 22, sand steppe vegetation, 17 Jun. 1999, J. Jeppson, M. Jeppson 4976 (GB)*. Ibidem, 17 Jun. 1997, S. Jeppson, J. Jeppson, M. Jeppson 3787 (GB)*; Orgovány, Orgományi rétek, Hollóshegy-Palinkás, sand steppe vegetation, 16 Jun. 1999, J. Jeppson, M. Jeppson 4966 (GB)*.

***Tulostoma* sp. 10**: HUNGARY, **Bács-Kiskun**: Kéleshalom, Kéleshalmi Borokás, sand steppe vegetation, 18 Jun. 1997, S. Jeppson, J. Jeppson, M. Jeppson 3813 (GB)*. SPAIN, **Huelva**: La Palma del Condado, Stipa grassland with Thymus, 1 Jan. 2003, S. Jeppson, J. Jeppson, M. Jeppson 6198 (GB)*.

***Tulostoma* sp. 11**: SPAIN, **Madrid**: Casa de Campo, 14 Nov. 1988, M. Jeppson 881114 (GB)*.

***Tulostoma* sp. 12**: ITALY, **Ravenna**: Pineta di Classe, sulla sabbia, su terreno erboso nella regione litorale, 13 Nov. 1992, I. Hausknecht (WU, AH 16793)*. SPAIN, **Girona**: Pals, en dunas, 21 Dec. 1986, J.M. Vidal 861221-17 (AH 15040, GB)*.

***Tulostoma* sp. 13**: USA, **New Mexico**: 4 mi. N. of Albuquerque, 30 May 1947, W.H. Long (as *T. excentricum*, herb. Long 11461, BPI 729350)*.

***Tulostoma* sp. 14**: SPAIN, **Zaragoza**: Caspe, Laguna Salada, in halophilous vegetation, 27 Apr. 1999, S. Jeppson, J. Jeppson, M. Jeppson 5004 (GB)*. Ibidem, M. Jeppson 5011 (GB)*.

***Tulostoma* sp. 15**: HUNGARY, **Bács-Kiskun**: Orgovány, Orgományi rétek, sand steppe vegetation, 17 Jun. 1999, J. Jeppson, M. Jeppson 990617b (GB)*. SPAIN, **Madrid**: Alcalá de Henares, cerros de Alcalá, gypsophilous steppe vegetation, at road verge, 2 Mar. 2011, G. Moreno, A. Altés, A. Stridvall, E. Larsson, M. Jeppson 9295 (GB, AH)*. Ibidem, 2 Mar. 2011, M. Jeppson 9296 (GB)*. Ibidem, Jan. 1993, G. Moreno, V. González, A. Altés (as *T. pseudopulchellum*, AH 15558)*.

***Tulostoma* sp. 16**: RUSSIAN FEDERATION, **Sakha**: Khangalasskiy Ulus, Oktyomtsy, steppe SW of village, 15 Aug. 1999, H. Knudsen 99.337 (C)*.

***Tulostoma* sp. 17**: SPAIN, **Madrid**: Alcalá de Henares, junto al cerro Malvecino, terreno margoso con algo de yeso, 7 Nov. 1990, equipo micología UAH (AH 13674, GB)*.

***Tulostoma* sp. 18**: SPAIN, **Ciudad Real**: Parque Nac. Tablas de Daimiel, 18 Mar. 1994, P. Juste (MA-F *35900*)*; **Zaragoza**, Bujaraloz, carretera 230, km 15, sheep-grazed steppe meadow, 6 Apr. 2010, E. Larsson. U. Andersson, T. Gunnarsson, T. Knutsson, M. Jeppson 9046 (GB)*.

***Tulostoma* sp. 19**: CYPRUS, 14 Feb. 2014, T. Læssøe, A. Molia 140115-2 (GB)*.

***Tulostoma* sp. 20**: SPAIN, **Zaragoza**: Los Monegros, Sástago, along road Escatrón-Sástago, 500 m N of Ebro-bridge, dry, sandy calcareous soil, 27 Apr. 1999, S. Jeppson, J. Jeppson, M. Jeppson 5015 (GB)*.

***Tulostoma* sp. 21**: SPAIN, **Madrid**: Alcalá de Henares, cerros de Alcalá, 3 Nov. 1989, (as *T. moravecii*, AH 11698)*.

Type specimens of species examined, from outside of Europe

Tulostoma albicans V.S. White: USA, **Texas**, 1893, E.D. Cope (holotype, NY)*. Figure 21.

Tulostoma egranulosum Lloyd: AUSTRALIA, **Victoria**, Melbourne: D. McAlpine (holotype, herb. Lloyd 15424, BPI 704420, GenBank KC333072).

Tulostoma excentricum Long: USA, **New Mexico**: E of Sandia Vista Court, Albuquerque, 20 Jun. 1941, W.H. Long (holotype, herb. Long 9360, BPI 729284)*.

Tulostoma leiosporum R.E. Fr.: ARGENTINA, **Jujuy**: Pampa Blanca, 23 May 1901, R. Fries 64 (lectotype, S).

Tulostoma lysocephalum Long: USA, **New Mexico**: on highway 70, 10 mi. W. of Deming, 12 Sept. 1941, W.H. Long, D.J. Stouffer (holotype, herb. Long 9639, BPI 749235)*. Figure 22.

Tulostoma macrocephalum Long: USA, **New Mexico**: White Sands Nat'l Monument, W of Administration Bldg, 22 Apr. 1942, W.H. Long (holotype, herb. Long 10113, BPI 729448)*.

Tulostoma readeri Lloyd: AUSTRALIA, Reader (holotype, herb. Lloyd 15421, BPI, GenBank KC333076).

Tulostoma rufum Lloyd: USA, **Alabama**: Spring Hill, A.S. Bertolet (lectotype, herb. Lloyd 25542, BPI 704578)*. Figure 23.

Tulostoma submembranaceum G. Moreno, C. Ochoa & J.E. Wright: MEXICO, **Baja California**: ad viam San Quintín-Cataviña km 35, 6 Dec. 1991, N. Ayala, C. Ochoa, F. Esteve-R., A. Altés, G. Moreno (holotype, AH 15132)*.

Tulostoma tuberculatum V.S. White: CANADA, **British Columbia**: Macoun (holotype, NY; isotype, herb. Long, BPI 729034, GenBank KC333070).

Tulostoma xerophilum Long: USA, **Arizona**: 7 mi from Nogales, on Hway 89, 13 Nov. 1936, Long & Stouffer (holotype, herb. Long 9688, BPI 802484)*. Figure 24.

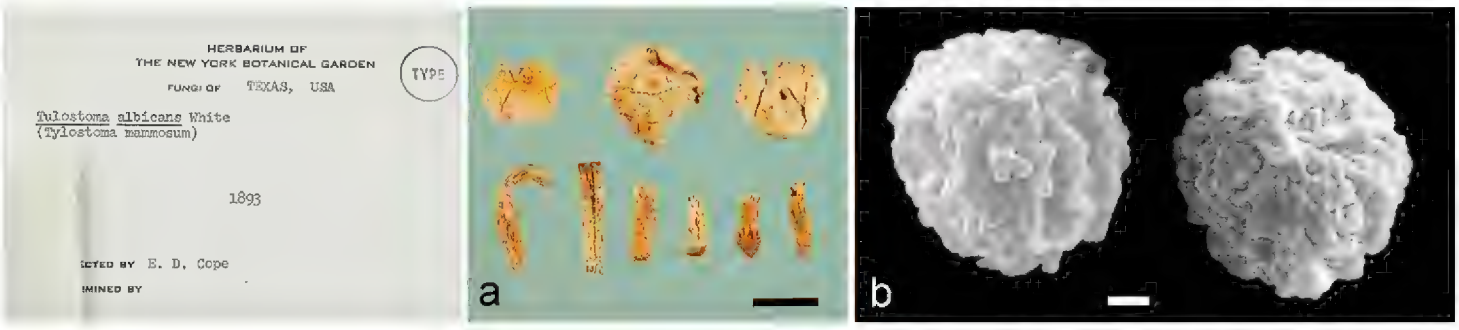

Figure 21. *T. albicans*, holotype (NY): a. basidiocarps **b** spore ornamentation under SEM. Scale bars: **a** = 10 mm; **b** = 1 µm.

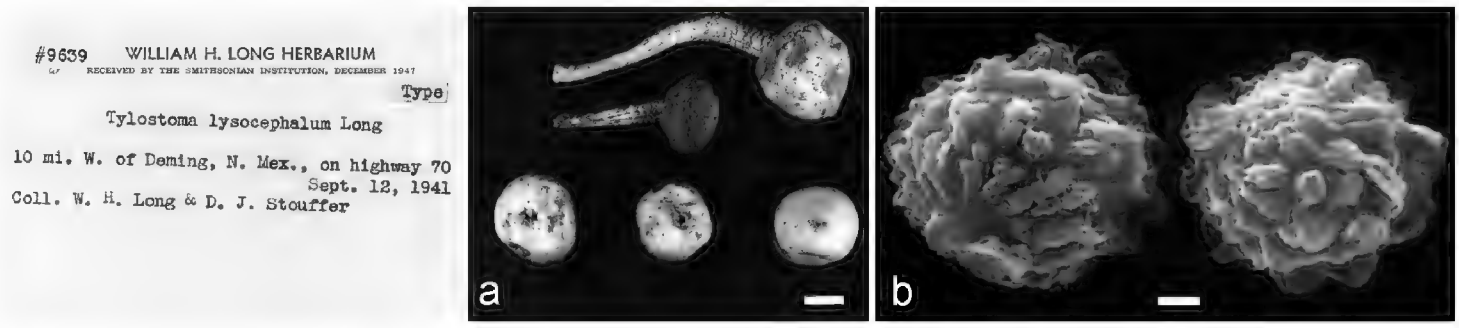

Figure 22. *T. lysocephalum*, holotype (herb. Long 9639, BPI 749235): **a** basidiocarps **b** spore ornamentation under SEM. Scale bars: **a** = 10 mm; **b** = 1 µm.

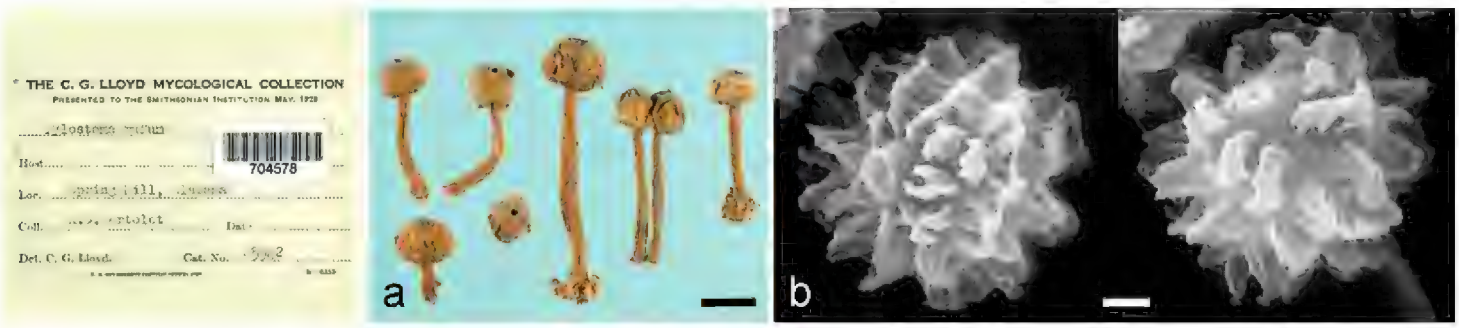

Figure 23. *T. rufum*, lectotype (herb. Lloyd 25542, BPI): **a** basidiocarps **b** spore ornamentation under SEM. Scale bars: **a** = 10 mm; **b** = 1 µm.

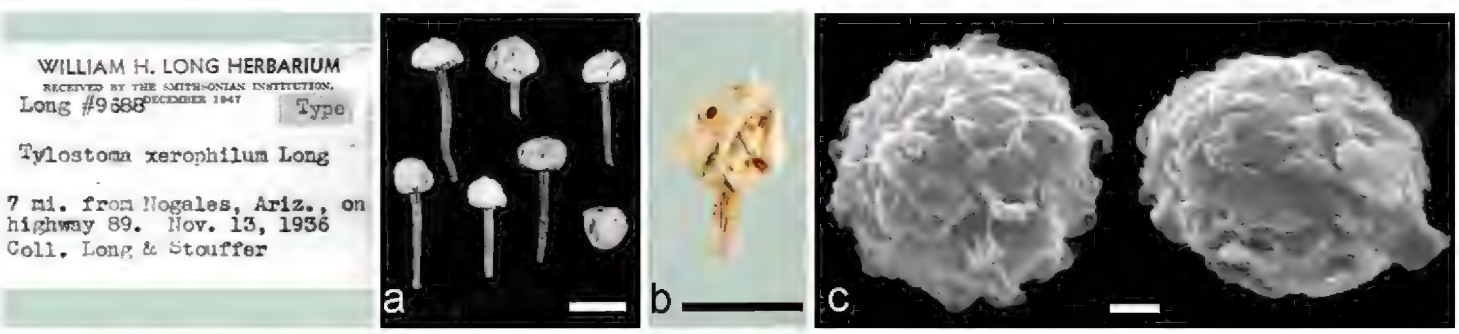

Figure 24. *T. xerophilum*, holotype (herb. Long 9688, BPI): **a–b** basidiocarps **c** spore ornamentation under SEM. Scale bars: **a–b** = 10 mm; **c** = 1 µm

Discussion

In this study, we found that the species diversity of *Tulostoma* was unexpectedly high, particularly from the arid areas of the world. The majority of the recovered 21 unidentified clades from the phylogenetic analysis (Figure 1a–c) comprise sequence data

from specimens collected in dry arid areas of Europe, notably Hungary and Spain. In Europe, the sand steppe vegetation of Hungary has been shown to be exceptionally species rich in gasteroid fungi (Hollós 1904, Rimóczi et al. 2011). The sand steppes of Hungary constitute a western outpost of the Euroasian steppe zone and in this study 14 of the included identified species of *Tulostoma* occur there, along with nine of the hithero unnamed species. Thus, we confirm the steppe vegetation as a species diversity hot spot for gasteroid fungi. Another identified area with high species diversity is the arid vegetation in the central part of the Iberian Peninsula, where 11 of the identified species and six of the hitherto unnamed species occur.

But species of *Tulostoma* are not restricted to arid regions only. Studies in the Tropics and temperate woodlands have indicated the presence of new species, growing in shaded, humus-rich, or even humid conditions (Hernández-Caffot et al. 2011). In Europe, there are also a few species that are adapted to grow in shaded and more humid conditions as *T. fulvellum*, *T. lloydii, T. niveum,* and *T. squamosum*.

In contrast to several recent phylogenetic studies of the Basidiomycota, where the traditional generic concepts of morphologically similar groups have shown to be polyphyletic and resulted in a number of new genera and higher taxa (e.g. Lodge et al. 2014, Örstadius et al. 2015, Miettinen et al. 2016, Petersen and Hughes 2016), *Tulostoma* is here confirmed as monophyletic. However, the recovered tree topology is not in congruence with the current infrageneric classification of *Tulostoma* (Pouzar 1958, Wright 1987), suggesting that many of the morphological characters used for delimitations are plesiomorphic or homoplasious. The endoperidial mouth region and the exoperidium, the major delimiting characters for the infrageneric classification when based on morphology, are here shown to be incongruent with the phylogeny, as the different types are distributed among the major clades. On the other hand, endo- and exoperidial features, as well as the spore morphology, are here confirmed and shown to be important characters for the delimitation of species.

Wright (1954) and Wright et al. (1972) listed morphological features in *Tulostoma* and classified their taxonomic importance as "primary" and "secondary". Wright (1987) divided the genus in two subgenera (*Tulostoma* and *Lacerata*), based on features of the mouth. The former subgenus was divided in two series (*Brumalia* and *Fimbriata*), each of them containing five sections. In our analyses Clade 3 corresponds more or less with the section *Fimbriata,* and a supported branch in Clade 9 corresponds partly to section *Poculata*. However, species belonging to Wright's sections *Brumalia* and *Hyphales* are distributed amongst the major clades and occur in Clades 3, 4, 5, 9, and 10 (Figure 1a–c). A new infrageneric classification of *Tulostoma* is necessary, but should be done in the light of further taxon sampling, sequence data, and analyses.

Several of the clades include groups with morphologically cryptic speciation, that are distinguished by the molecular data, but for which discriminatory morphological features are ambiguous or lacking. This is particularly evident in Clades 2, 3, 5, and 10, where a number of unidentified taxa from xerothermic habitats were recovered.

The phylogenetic analyses recovered and confirmed the classical, well-known species from Europe, namely *T. brumale, T. cyclophorum, T. fimbriatum, T. fulvellum, T.*

giovanellae, T. kotlabae, T. lloydii, T. melanocyclum, T. pulchellum, and *T. squamosum*. In addition, one species not previously known from the continent – *T. punctatum* – was identified. Further *Tulostoma calcareum, T. calongei* and *T. eckbladii* are described as new to science. The former has a wide distribution in Europe whereas the two latter are hitherto only known from Central Spain and northernmost Norway respectively.

We also discovered species with misapplied names and two species new to science were identified from the continental sand steppe vegetation in East Central Europe: *T. grandisporum* and *T. pannonicum*. These had previously been misinterpreted as *T. leiosporum* (Jeppson et al. 2011, Rimóczi et al. 2011). The name *T. obesum* has in current use been applied for a species found in dry sandy habitats in East Central Europe as well as in dry steppic sites on the Iberian Peninsula (Altés et al. 1999, Rimóczi et al. 2011). It is characterized by having smooth spores and a large, whitish spore-sac that opens lacerately at maturity. In this study an ITS sequence from of the isotype of *T. obesum* (from Colorado, USA) was generated and included in the analyses. It did not form a clade with the European concept of *T. obesum* but with a hitherto unnamed species in Clade 2, known from sand dune habitats of SE Spain. Thus we here confirm *T. obesum* from Europe, but with a different species interpretation from that of previous European mycologists (Altés et al. 1999, Rimóczi et al. 2011).

The Hungarian and Spanish collections, previously attributed to *T. obesum*, are placed in Clade 6 and are here treated as *T.* aff. *cretaceum*. This clade consists of a complex of closely related cryptic species with a strong geographical signal. In East Central Europe and Central Asia the name *T. volvulatum* I.G. Borshch. has been misapplied to cover this species complex (e.g. Hollós 1904, Shvartsman and Filimonova 1970). As demonstrated by Altés et al. (1999), the holotype of *T. volvulatum* was based on a specimen of *T. giovanellae,* with typical ornamented spores. Under the name *T. volvulatum*, Hollós (1904) recorded *T. giovanellae* (e.g. S, F265540) but also specimens with smooth spores (Babos 1999) that closely match Sorokin´s (1890) interpretation of *T. volvulatum* from the Karakoum Desert (Sorokin 1890, plate XXV, Figure 353a). A number of poorly known species described from desert habitats in northern Africa (e.g. *T. barbeyanum* Henn., *T. boissieri* Kalchbr., *T. giolianum* Bacc., and *T. ruhmerianum* Henn.) as well as the American *T. cretaceum* Long and *T. meristostoma* Long, share characters typical of this clade and need to be included in an extended study, which should also deal with several recently described species from China (*T. brevistipitatum* B. Liu, Z.Y. Li & Du, *T. sabulosum* B. Liu, Z.Y. Li & Du, and *T. helanshanense* B. Liu, Z.Y. Li & Du). Calonge (1998) identified Spanish material attributed to *T. volvulatum* as *Schizostoma laceratum* (Ehrenb.: Fr.) Lév. Based on sequence data, Gube (2009) recovered *S. laceratum* as closely related to the type of *T. cretaceum*. As pointed out by Kuhar et al. (2012), the whole complex is in need of a detailed study comprising morphological as well as molecular data. Similarly, the status of the genus *Schizostoma* is unclear and calls for further study.

Tulostoma simulans is here identified from Europe. According to our data it seems to have a wide distribution but appears to be rare. The species is known from East Central Europe under the name *T. moravecii*, and it may have been overlooked due to its resemblance with *T. brumale*.

The application of the name *Tulostoma caespitosum* Trab. is an unsolved problem. The species was described from northern Africa and later reported from the Americas, Asia, and Europe (Calonge 1998, Calonge and Wright 1988, Wright 1987). Among its main characteristics, according to Wright (1987), are the distorted basidiomata and the sometimes caespitose growth. Wright (1987) did not find any collection that he considered to be suitable as a holotype and suggested a neotype, collected by Trabut in Algeria, coinciding with the protologue of *T. caespitosum* (*T. caespitosum* var. *acaule*, herb. Patouillard 1422, FH; unpublished herbarium name). However, Wright did not formally designate a neotype. An authentic collection of *T. caespitosum* var. *acaule*, from Algeria (leg. Trabut), is kept in BPI (ex herb. Patouillard, herb. Lloyd 15427, BPI 704324) but was unfortunately found to be in poor condition and too scanty for DNA sequencing. A second collection from Africa (indet. region) in BPI (Patouillard 4639, herb. Lloyd 15426, BPI 704323) was sequenced for the present study and found to be identical with *T. giovanellae*, in Clade 10, Figure 1c. The spore ornamentation of this specimen is identical to *T. giovanellae*. However, in collection BPI 704324 the spores were shown to be strongly verrucose, thus differing from *T. giovanellae*. The typification of *T. caespitosum* thus remains unsettled and the unclear interpretation of this name calls for further study.

Kreisel and Al-Fatimi (2008) observed that gasteroid basidiomycetes from desert habitats tend to produce smooth spores. This stands in contrast to taxa growing in vegetation types that are regularly exposed to rainfall – these usually have more or less distinctly ornamented, verrucose–echinate spores. Kuhar et al. (2012) found that the relative abundance of species with smooth spores within the study in Argentina reached a maximum under desert conditions. The relative abundance of species with ornamented spores was shown to go up with increasing humidity. Their hypothetical explanation was that the spore wall ornamentation increases the water repellency, favouring a wind-blown dispersal (Kreisel and Al-Fatimi 2008).

In this study, species of *Tulostoma* with smooth or sub-smooth spores are found to occur mainly in arid, and exposed habitats in East Central Europe and Spain (*T.* aff. *cretaceum, T. grandisporum, T. obesum, T. pannonicum*). Species with moderately to strongly ornamented spores (*T. beccarianum, T. brumale, T. calongei, T. giovanellae, T. melanocyclum, T. punctatum, T. simulans,* and *T. subsquamosum*) are found in more steppic, dry grassland communities in southern and east central Europe. In semi-shaded or shaded to somewhat humid habitats, the genus is represented by species with ornamented spores (*T. cyclophorum, T. niveum,* and *T. squamosum*) but also a few with completely smooth spores (*T. fulvellum* and *T. lloydii*). Among the species regularly encountered in northern and north-western Europe, i.e. in sub-oceanic or oceanic climates, all have moderately to distinctly ornamented spores (*T. eckbladii, T. brumale, T. calcareum, T. fimbriatum, T. kotlabae, T. melanocyclum, T. niveum, T. simulans, T. squamosum,* and *T. winterhoffii*). In accord with the observations by Kreisel and Al-Fatimi (2008), *Tulostoma* species with smooth or sub-smooth spores tend to occur mainly in dry and arid habitats whereas those with distinct and strongly ornamented spores grow in more humid habitats.

Due to difficulties to determine *Tulostoma* specimens to species level, many published records and herbarium collections are based on misinterpretations. It is therefore problematic to evaluate species distribution data. Some seem to have wide, more or less cosmopolitan distribution ranges (e.g. *T. fimbriatum, T. pulchellum, T. striatum, T. squamosum,* and *T. subsquamosum*). Others appear to be restricted to certain geographical areas or habitat types (e.g. *T. grandisporum, T. pannonicum, T. niveum,* and *T. eckbladii*). In Europe most species have southern and southeastern distributions. The number of species decreases towards the north. *Tulostoma brumale, T. calcareum, T. fimbriatum, T. kotlabae, T. melanocyclum, T. simulans,* and *T. winterhoffii* reach the south of Fennoscandia, whereas *T. niveum* is regularly found between 55° and 62° N, from southern Finland westwards to northern Scotland. The report herein of *T. niveum* from near Geneva in Switzerland is a significant range extension. The newly described species, *T. eckbladii*, is, as far as currently known, limited to areas above the Arctic Circle.

A number of species of *Tulostoma* occur in habitat types that are under the threat of changing agricultural methods, anthropogenic exploitation, and construction works. Hernández-Caffot et al. (2011) described *T. domingueziae* from threatened woodland ecosystems in Argentina. In Europe, several *Tulostoma* species are on the red-list of threatened species in the countries where they occur: *T. brumale, T. fimbriatum, T. kotlabae, T. melanocyclum, T. simulans (*as *T. moravecii), T. pulchellum,* and *T. niveum* (ArtDatabanken 2015, ArtsDatabanken 2015, Holec and Beran 2006, Rassi et al. 2010). For *T. niveum*, national action plans for its conservation have been elaborated and become effective in Great Britain and Sweden (BAP 2005, Jeppson 2005, 2006b). *Tulostoma niveum* has also been nominated for the IUCN global red-list (http://iucn.ekoo.se/iucn/species_view/325133) and *T.* aff. *cretaceum* (as *T. volvulatum*) is legally protected in Hungary (Siller et al. 2005). Many species of *Tulostoma* can be considered as indicators of valuable and threatened habitats.

In this study we show, based on molecular data, that the species diversity of *Tulostoma* is much higher than previously known. Much of this detected diversity is associated with dry, arid areas, such as the steppe vegetation in Hungary. This suggests that the number of species is likely to increase even more as other dry areas of the world are explored.

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References

Altés A, Moreno G (1991) *Tulostoma striatum* (Gasteromycetes, Basidiomycotina) new for Europe. Cryptogamie Mycologie 12(2): 149–153.

Altés A, Moreno G (1993) *Tulostoma beccarianum* Bresad., the correct name for *T. simulans* Lloyd. Mycotaxon 48: 223–227.

Altés A, Moreno G, Hausknecht A (1994) *Tulostoma kotlabae* Pouzar in Italia. Rivista di Micologia 38(2): 167–172.

Altés A, Moreno G (1995) *Tulostoma fimbriatum*, the correct name for *Tulostoma readeri*. Mycotaxon 56: 421–425.

Altés A, Moreno G, Wright JE (1996) New data on *Tulostoma subsquamosum* (Gasteromycetes). Cryptogamie Mycologie 17(30-4): 139–148.

Altés A, Moreno G (1999) Notes on type materials of *Tulostoma* (Tulostomataceae) *T. macrosporum*, *T. meridionale*, *T. utahense*. Persoonia 17(2): 259–264.

Altés A, Moreno G, Wright JE (1999) Notes on *Tulostoma volvulatum* and *T. giovanellae*. Mycological Research 103(1): 91–98. https://doi.org/10.1017/S0953756298006704

Antonín V, Kreisel H (2008) *Tulostoma leslei*, a new record from Italy. Österreichische Zeitschrift für Pilzkunde 17: 81–85.

Ariyawansa HA, Hawksworth DL, Hyde KD, Jones EBG, Maharachchikumbura DSM, Manamgoda DS, Thambugala KM, Udayanga D, Camporesi E, Daranagama A, Jayawardena R, Liu J-K, McKenzie EHC, Phookamsak R, Senanayake IC, Shivas RG, Tian Q, Xu J-C (2014) Epitypification and neotypification: Guidlines with appropriate and inappropriate examples. Fungal Diversity 69(1): 57–91. https://doi.org/10.1007/s13225-014-0315-4

ArtDatabanken (2015) Rödlistade arter i Sverige 2015. ArtDatabanken, SLU, Uppsala. Available at http://www.artdatabanken.se

ArtsDatabanken (2015) Norsk rødliste for arter 2015. Available at http://data.artsdatabanken.no/Rodliste

Asai I (2004) *Tulostoma striatum* in Japan [in Japanese]. Nippon Kingakkai Kaiho 45: 11–13.

Asai I, Asai Y (2008) New records of *Tulostoma* species from Japan. Mycoscience 49(6): 399–402. https://doi.org/10.1007/s10267-008-0430-8

Babos M (1999) Higher fungi of Basidiomycetes. of the Kiskunság Nemzeti Park and its environs. In: Lökös L, Rajczy M (Eds) The flora of the Kiskunság National Park, II. Magyar Természettudomány Múzeum, Budapest, 199–299.

BAP (2005) Action Plan for *Tulostoma niveum*. Biodiversity Action plan. http://www.ukbap.org.uk/ukplans.aspx?ID=620

Baseia IG, Milanez AI (2002) *Tulostoma* Persoon (Gasteromycetes) from the Cerrado region, state of São Paulo, Brazil. Acta Botanica Brasilica 16(1): 9–14. https://doi.org/10.1590/S0102-33062002000100003

Bataille F (1910) Champignons rares ou nouveaux de la Franche-Comté. Bulletin de la Société Mycologique de France 26(1): 138–149.

Bohus G, Babos M (1977) Fungorum rariorum icones coloratae VIII. J. Cramer, Vaduz.

Bottomley AM (1948) Gasteromycetes of South Africa. Bothalia 4(3): 473–810. https://doi.org/10.4102/abc.v4i3.1859

Brochmann C, Rustan Ø, Brandrud TE (1981) Gasteromycetslekten *Tulostoma* styltesopp. i Norge. Blyttia 39: 209–226.

Candoussau F (1973) Champignons rares à Vitoria (Espagne). Bulletin de la Société Mycologique du Béarn 53: 3–6.

Calonge FD (1992) Nuevos datos sobre el género *Tulostoma* (Gasteromycetes) en España. *Tulostoma bruchi* Spegaz., nuevo para Europa. Bulletin de Sociedad Micologica de Madrid 16: 151–153.

Calonge FD (1998) Gasteromycetes, I. Lycoperdales, Nidulariales, Phallales, Sclerodermatales, Tulostomatales. Flora Mycologica Iberica Vol. 3. Real Jardín Botánico Madrid. J. Cramer. Madrid, Berlin, Stuttgart.

Calonge FD (2000) Validation and confirmation of some new taxa recently published. Bulletin de Sociedad Micologica de Madrid 25: 301–302.

Calonge FD, Almeida MG (1992) *Tulostoma lusitanicum* sp. nov. (Gasteromycetes). Bulletin de Sociedad Micologica de Madrid 16: 109–113.

Calonge FD, Llimona X, Martín MP (2007) Nuevos datos sobre el género *Tulostoma* (Gasteromycetes) en España IV. Revista Catalana de Micología 29: 11–16.

Calonge FD, Martín MP (1992) Nuevos datos sobre el género *Tulostoma* (Gasteromycetes) en España II. *Tulostoma xerophilum*, nueva para Europa. Bulletin de Sociedad Micologica de Madrid 17: 115–122.

Calonge FD, Wright JE (1988) El género *Tulostoma* Pers.: Pers. (Gasteromycetes) en España. Bulletin de Sociedad Micologica de Madrid 13: 119–138.

Clark K, Karsch-Mizrachi I, Lipman DJ, Ostell J, Sayers E (2016) Genbank. Nucleic Acids Research 44(D1): 67–72. https://doi.org/10.1093/nar/gkv1276

Coetzee JC (2007) Key to the genus *Tulostoma* in southern Africa. Extracted and adapted from Wright (1987). Mycoafrica 3(2): 4–8.

Coker WC, Couch JN (1928) The Gasteromycetes of the eastern United States and Canada. The University of North Carolina Press, 201 pp. [Reprint 1974, Dover Publications Inc. New York]

Cortez VG, Baseia IG, Silveira RM (2009) Gastroid mycobiota of Rio Grande do Sul, Brazil: Tulostomataceae. Mycotaxon 108: 373–392. https://doi.org/10.5248/108.365

Cunningham GH (1925) The Gasteromycetes of Australasia II. A revision of *Tulostoma*. Proceedings of The Linnean Society of New South Wales 50: 245–258.

Cunningham GH (1932) The gasteromycetes of Australasia XIV. The family Tulostomataceae. Proceedings of The Linnean Society of New South Wales 57: 27–38.

Demoulin V (1984) *Tulostoma cyclophorum* C. Lloyd (Syn.: *T. pampeanum* (Speg.) Wright), gastéromycète méconnu en Europe et dans le bassin méditerranéen. Bulletin trimestriel de la Société mycologique de France 100(2): 207–210.

Dörfelt H, Bumžaa D (1986) Die Gasteromyceten (Bauchpilze) der Mongolischen Volksrepublik. Nova Hedwigia 43(1–2): 87–111.

Dörfelt H, Täglich U (1990) Pilzfloristische Arbeitsergebnisse aus der Mongolischen Volksrepublik. Boletus 14(1): 1–27.

Drummond AJ, Suchard MA, Xie D, Rambaut A (2012) Bayesian Phylogenetics with BEAUti and the BEAST 1.7. Molecular Biology and Evolution 29(8): 1969–1973. https://doi.org/10.1093/molbev/mss075

Eckblad FE (1951). The genus *Tulostoma* Pers. in Norway. Blyttia 9:33–36.

Esqueda M, Moreno G, Perez-Silva E, Sánchez A, Altés A (2004) The genus *Tulostoma* in Sonora, Mexico. Mycotaxon 90(2): 409–422.

Fries EM (1829) Systema Mycologicum III. Gryphiswaldae.

Fries EM (1849) Summa Vegetabilium Scandinaviae. Sectio posterior. Upsaliae.

Gardes M, Bruns TD (1993) ITS primers with enhanced specificity for basidiomycetes-application to the identification of mycorrhizas and rusts. Molecular Ecology 2(2): 113–118. https://doi.org/10.1111/j.1365-294X.1993.tb00005.x

Gube M (2009) Ontogeny and phylogeny of gasteroid members of Agaricaeae (Basidiomycetes). Thesis, Friedrich-Schiller-Universität, Jena. https://www.db-thueringen.de/servlets/MCRFileNodeServlet/dbt_derivate_00019285/Gube/Dissertation.pdf

Hernández-Caffot ML, Dominguez LS, Hosaka K, Crespo EM (2011) *Tulostoma domingueziae* sp. nov. from *Polylepis australis* woodlands in Córdoba Mountains, central Argentina. Mycologia 103(5): 1047–1054. https://doi.org/10.3852/10-266

Hernández-Navarro E, Gutierrez A, Barredo-Pool F, Esqueda M (2015) Especies de *Tulostoma* (Basidiomycetes, Agaricomycetes) en un matorral espinoso de Sonora, México. Revista Mexicana de Micologia 41: 65–72.

Hibbett DS, Pine EM, Langer E, Langer G, Donaghue MJ (1997) Evolution of gilled mushrooms and puffballs inferred from ribosomal DNA sequences. Proceedings of the National Academy of Sciences of the United States of America 94(22): 12002–12006. https://doi.org/10.1073/pnas.94.22.12002

Holec J, Beran M (2006) Červený seznamn hub (makromycetů) České republiky. Příroda 24: 1–280.

Hollós L (1904) Die Gasteromyceten Ungarns. Oswald Weigel, Leipzig.

Hollós L (1913) Zu den Gasteromyceten Ungarns – Magyarország Gasteromycetái-hoz. Magyar Botanikai Lapok 12(6–7): 194–200.

Hopple JS Jr, Vilgalys R (1999) Phylogenetic relationships in the mushroom genus *Coprinus* and dark spored allies based on sequence data from the nuclear gene coding for the large

ribosomal subunit RNA: divergent domains, outgroups, and monophyly. Molecular Phylogenetics and Evolution 1381: 1–19. https://doi.org/10.1006/mpev.1999.0634

Hussain S, Yousaf N, Afshan N, Niazi AR, Ahmad H, Khalid AN (2016) *Tulostoma ahmadii* sp. nov. and *T. squamosum* from Pakistan. Turkish Journal of Botany 40: 218–225. https://doi.org/10.3906/bot-1501-9

Jeppson M (2005) Åtgärdsprogram för bevarande av vit stjälkröksvamp (*Tulostoma niveum*) Naturvårdsverket. Rapport 5512. https://www.naturvardsverket.se/Documents/publikationer/620-5512-7.pdf

Jeppson M (2006a) Gråhvid Stilkbovist (*Tulostoma kotlabae*) ny for Danmark. Svampe 54: 9–13.

Jeppson M (2006b) *Tulostoma niveum* – en av världens sällsyntaste svampar. Svensk Mykologisk Tidskrift 27(2): 58–63.

Jeppson M (2008) The genus *Tulostoma* in Slovakia. Catathelasma 10: 5–19.

Jeppson M, Benedek L, Rimóczi I (2011) Három új pöfetegféle a hazai fungában – Three gastroid species new to the Hungarian mycobiota. Mikológiai Közlemények 50(1): 107–118.

Kabát V (1997) Nález vzácneho stopkatca hladkovýtrusného *Tulostoma fulvellum* Bres. in Petri. Spravodajca 5(16): 10–11.

Kreisel H (1984) Karten der Pflanzenverbreitung in der DDR. 6. Serie. Die Stielboviste (Gattung *Tulostoma*) der Deutschen Demokratischen Republik und Westberlins. Hercynia 21: 396–416.

Kreisel H (2001) Checklist of the gasteral and secotioid Basidiomycetes of Europe, Africa and the Middle East. Österreichische Zeitschrift für Pilzkunde 10: 213–313.

Kreisel H (2004) *Tulostoma pulchellum* in Brandenburg – neu für Deutschland. Zeitschrift für Mykologie 70(1): 107–110.

Kreisel H, Al-Fatimi M (2008) Further Basidiomycetes from Yemen. Feddes Repertorium 119(5–6): 463–483. https://doi.org/10.1002/fedr.200811175

Kreisel H, Scholler M (2008) Funde von Stielbovisten (*Tulostoma*) jenseits des Mittelmeeres. Carolinea 66: 71–76.

Kříž M (2011) New finds of *Tulostoma pulchellum* in Bohemia. Mykologické Listy 115: 7–15.

Kuhar F, Castiglia V, Zamora JC, Papinutti L (2012) New records and notes on gasteroid fungi of arid regions in Argentina. Sydowia 64(2): 233–244.

Larsson A (2014) AliView: a fast and lightweight alignment viewer and editor for large data sets. Bioinformatics 30(22): 3276–3278. https://doi.org/10.1093/bioinformatics/btu531

Larsson E, Jacobsson S (2004) The controversy over *Hygrophorus cossus* settled using ITS sequence data from 200-year-old type material. Mycological Research 108(7): 781–786. https://doi.org/10.1017/S0953756204000310

Larsson E, Örstadius L (2008) Fourteen coprophilous species of *Psathyrella* identified in the Nordic countries using morphology and nuclear rDNA sequence data. Mycological Research 112(10): 1165–1185. https://doi.org/10.1016/j.mycres.2008.04.003

Linnaeus C (1753) Species plantarum. Holmiae.

Liu B (1979) Two new species of *Tulostoma* from China. Mycologia 71(6): 1275–1278. https://doi.org/10.2307/3759119

Lloyd CG (1906) The Tylostomae. Mycological Writings 2: 9–28, plates 74–85. Cincinnati, USA.

Lodge DJ, Padamsee M, Matheny PB, Aime MC, Cantrell SA, Boertmann D, Kovalenko A, Vizzini A, Dentinger BTM, Kirk PM, Ainsworth AM, Moncalvo J-M, Vilgalys R, Larsson E, Lücking R. Griffith GW, Smith ME, Norvell LL, Desjardin DE, Redhead SA, Ovrebo CL, Lickey E.B, Ercole E, Hughes KW, Courtecuisse R, Young A, Binder M, Minnis AM, Lindner DL, Ortiz-Santana B, Haight J, Læssøe T, Baroni TJ, Geml J, Hattori T (2014) Molecular phylogeny, morphology, pigment chemistry and ecology in Hygrophoraceae (Agaricales). Fungal Diversity 64(1): 1–99. https://doi.org/10.1007/s13225-013-0259-0

Long WH (1944) Studies in the Gasteromycetes X. Seven new species of *Tulostoma*. Mycologia 36(4): 318–339. https://doi.org/10.2307/3754748

Long WH (1946) Studies in the Gasteromycetes XII. Five species of *Tylostoma* with membranous exoperidia. Mycologia 38(1): 77–90. https://doi.org/10.2307/3755049

Long WH, Ahmad S (1947) The genus *Tylostoma* in India. Farlowia 3(2): 225–267.

Martín MP (1990) Notas sobre Gasteromicetes I: *Tulostoma fimbriatum* var. *heterosporum* J.E. Wright. Butlletí Societat Catalana de Micologia 13: 61–65.

Martín MP, Calonge FD (1994) Nuevos datos sobre el genero *Tulostoma* (Gasteromycetes) en España III. Butlletí Societat Catalana de Micologia 16–17: 205–212.

Matheny PB, Curtis JM, Hofstetter V, Aime MC, Moncalvo JM, Ge ZW, Yang ZL, Slot JC, Ammirati JF, Baroni TJ, Bougher NL, Hughes KW, Lodge DJ, Kerrigan RW, Seidl MT, Aanen DK, DeNitis M, Daniele GM, Desjardin DE, Kropp BR, Norvell LL, Parker A, Vellinga EC, Vilgalys R, Hibbett DS (2006) Major clades of Agaricales: a multilocus phylogenetic overview. Mycologia 98(6): 982–995. https://doi.org/10.3852/mycologia.98.6.982

Miettinen O, Spirin V, Vlasák J, Rivoire B, Stenroos S, Hibbett D (2016) Polypores and genus concepts in Phanerochaetaceae (Polyporales, Basidiomycota). MycoKeys 17: 1-46. https://doi.org/10.3897/mycokeys.17.10153

Monthoux O, Röllin O (1974) La flore fongique des stations xériques de la region de Genève. I. Introduction et Tulostomatales. Candollea 29: 309–325.

Moreno G, Altés A (1992) *Tulostoma simulans* (Gasteromycetes), una especie generalmente mal interpretada en España. Boletín de la Sociedad Argentina de Botánica 28(1–4): 159–164.

Moreno G, Altés A, Ayala N (1990) *Tulostoma cyclophorum* Lloyd (Gasteromycetes, Basidiomycotina) Nuevo para España peninsular. Anales del Instituto de Biologia, Serie Botanica 60(1): 21–25.

Moreno G, Altés A, Hausknecht A (1995a) *Tulostoma opacum* Long in northern Africa. Mycotaxon 54: 179–182.

Moreno G, Altés A, Ochoa C, Wright JE (1995b) Contribution to the study of the Tulostomataceae in Baja California, Mexico. I. Mycologia 87(1): 96–120. https://doi.org/10.2307/3760953

Moreno G, Altés A, Ochoa C, Wright JE (1997) Notes on type materials of *Tulostoma*. Some species with mixed holotypes. Mycolological Research 101(8): 957–965. https://doi.org/10.1017/S0953756297003572

Moreno G, Altés A, Wright JE (1992a) *Tulostoma squamosum*, *T. verrucosum* and *T. mussooriense* are the same species. Mycotaxon 43: 61–68.

Moreno G, Altés A, Wright JE (1992b) *Tulostoma pseudopulchellum* sp. nov. (Tulostomatales, Gasteromycetes) and allied species. Mycotaxon 43: 479–486.

Moreno G, Horak E, Altés A (2002) *Tulostoma kreiselii*, a new species for Tunisia, northern Africa. Feddes Repertorium 113(1–2): 7–10. https://doi.org/10.1002/1522-239X(200205)113:1/2<7::AID-FEDR7>3.0.CO;2-5

Morgan AP (1891) North American fungi. Third paper. The Gasteromycetes. The Journal of the Cincinnati Society of Natural History 12: 163–172.

Mrazek E, Krisai-Greilhuber I, Hausknecht A (1995) Bemerkenswerte epigäische Gasteromyzeten-Funde in Ostösterreich. Österreichische Zeitschrift für Pilzkunde 4: 11–33.

Nilsson RH, Tedersoo L, Abarenkov K et al. (2012) Five simple guidelines for establishing basic authenticity and reliability of newly generated fungal ITS sequences. MycoKeys 4: 37–63. https://doi.org/10.3897/mycokeys.4.3606

Nilsson RH, Wurzbacher C, Bahram M et al. (2016) Top 50 most wanted fungi. MycoKeys 12: 29–40. https://doi.org/10.3897/mycokeys.12.7553

Nitare J (1988) Stjälkröksvampen *Tulostoma kotlabae* i Sverige. Jordstjärnan 9(2): 7–10.

Nitare J (1997) *Tulostoma squamosum*. In: Larsson K-H (ed) Rödlistade svampar i Sverige – Artfakta. ArtDatabanken, SLU, Uppsala, 528 pp.

Nylander JAA (2004) MrModeltest v2. Program distributed by the author.

Örstadius L, Ryberg M, Larsson E (2015) Molecular phylogenetics and taxonomy in Psathytellaceae (Agaricales) with focus on psathyrelloid species: introduction of three new genera and 18 new species. Mycological Proggress 14: 25. https://doi.org/10.1007/s11557-015-1047-x

Peck CH (1896) New species of fungi. Bulletin of the Torrey Botanical Club 23(10): 411–420. https://doi.org/10.2307/2477789

Persoon CH (1794) Neuer Versuch einer systematischen Eintheilung der Schwämme. Neues Magazin für die Botanik 1: 63–128.

Persoon CH (1801) Synopsis Methodica Fungorum. Gottingae.

Petersen RH, Hughes KW (2016) Micromphale sect. Perforantia (Agaricales, Basidiomycetes); Expansion and phylogenetic placement. MycoKeys 18: 1–122. https://doi.org/10.3897/mycokeys.18.10007

Petri L (1904) Sul valore diagnostic del capillizio nel genere "*Tylostoma*" Pers. Annales Mycologici 2(5): 412–438.

Petri L (1909) Gasterales. Flora Italica Cryptogama. Fungi: 5. Società Botanica Italiana, 139 pp.

Piña C, Esqueda M, Altés A, Gutiérrez A (2010) First record of *Tulostoma gracilipes* (Agaricales, Agaricaceae) for the Americas. Mycotaxon 113: 371–376. https://doi.org/10.5248/113.371

Pouzar Z (1958) *Tulostoma* Pers. ex Pers. – Palečka. In: Pilát (Ed.) Gasteromycetes – Houbybřichatky. Flora ČSR, B-1. Praha, 589–613.

Rassi P, Hyvärinen E, Juslén A, Mannerkoski I (Eds) (2010) Suomen lajien uhanalaisuus punanen kirja 2010 – The 2010 red list of Finnish species. Ministry of the Environment. Finnish Environment Institute. Helsinki.

Rebriev YA, Gorbunova IA (2007) Gasteromitsety yoga zapadnoy i sredney Sibir. Sibirskiy Botanicheskaya Elektronny Zhurnal 1(1): 51–60.

Rehner SA, Buckley E (2005) A Beauviera phylogeny inferred from nuclear ITS and EF1-α sequences: evidence for cryptic diversification and links to Cordyceps teleomorphs. Mycologia 97(1): 84–98.

Rimóczi I, Jeppson M, Benedek L (2011) Characteristic and rare species of Gasteromycetes in Eupannonicum. Fungi non delineati 56–57. Edizioni Candusso, Alassio.

Ronquist F, Teslenko M, van der Mark P, Ayres DL, Darling A, Höhna S, Larget B, Liu L, Suchard MA, Huelsenbeck JP (2012) MrBayes 3.2, efficient Bayesian phylogenetic inference and model choice across a large model space. Systematic Biology 61(3): 539–542. https://doi.org/10.1093/sysbio/sys029

Rudnicka-Jezierska W (1991) Grzyby (Mycota) XXII. Flora Polska. Polska Akademia Nauk, Kraków.

Sarasini M (2003) Alcune rare Tulostomataceae dei litorali mediterranei. Rivista di Micologia 46(1): 7–14.

Sarasini M (2005) Gasteromiceti Epigei. A.M.B. Fondazione Centro Studi Micologici, Vicenza.

Schroeter J (1876) Ueber die Entwicklung und die systematische Stellung von *Tulostoma* Pers. In: Cohn's Beiträge zur Biologie d. Pflanzen 2: 65–71.

Schubert H, Specht P (2013a) Seltene Gasteromyceten in Deutschland III. *Tulostoma*: weitere Neufunde und Ergänzungen zur Verbreitung der Gattung in Deutschland und Europa. Zeitschrift für Mykologie 79(2): 405–416.

Schubert H, Specht P (2013b) Seltene Gasteromyceten in Deutschland IV – *Tulostoma winterhoffii*, ein neuer Stielbovist. Zeitschrift für Mykologie 79(2): 417–430.

Sesli E, Wright JE,Türkekul I (2000) The genus *Tulostoma* Pers.: Pers. (Gasteromycetes) in Turkey. Turkish Journal of Botany 24: 269–271.

Shvartsman SR, Filimonova NM (1970) Gasteromitsety – Gasteromycetes. Flora Sporovykh Rasteniy Kazakhstana VI. Akademiya Nauk Kazakhskoy SSR. Alma-Ata.

Siller I, Vasas G, Pál-Fám F, Bratek Z, Zagyva I, Fodor L (2005) Hungarian distribution of the legally protected macrofungi species. Studia Botanica Hungarica 36: 131–163.

Silva BDB (2006) Diversidade de Tulostomataceae Fischer (Gasteromycetes) em algunas áreas dos Estados do Rio Grand do Norde, Paraíba e Pernambuco. Dissertação de Mestrado, Universidade Federal do Rio Grande do Norte.

Silva BDB, Calonge FD, Baseia IG (2007) Studies in *Tulostoma* (Gasteromycetes) in the Neotropics. Some Brazilian species. Mycotaxon 101: 47–54.

Smith AM (1951) Puffballs and their allies in Michigan. University of Michigan Press. Ann Arbor. 131 pp.

Sorokin N (1890) Materiaux pour la flore cryptogamique de l'Asie Centrale. Revue de Mycologie 12: 49–69.

Specht P, Schubert H (2012) Seltene Gasteromyceten in Deutschland II. *Tulostoma pulchellum* var. *pulchellum*. Zeitschrift für Mykologie 78(2): 179–202.

Specht P, Schubert H, Lode D (2016) Seltene Gasteromyceten aus Deutschland (V) – *Tulostoma palatinum*, ein neuer Stielbovist. Zeitschrift für Mykologie 82(1): 65–84.

Swofford DL (2003) PAUP*. Phylogenetic Analysis Using Parsimony (* and Other Methods). Version 4. Sunderland, Massachusetts: Sinauer.

Tedersoo L, Anslan S, Bahram M et al. (2015) Shotgun metagenomes and multiple primer pair-barcode combinations of amplicons reveal biases in metabarcoding analyses of fungi. MycoKeys 10: 1-43. https://doi.org/10.3897/mycokeys.10.4852

Tomaszewska A, Łuszczyński J, Łuczyńska B, Jaworska J (2011) Taxonomic notes on the Polish *Tulostoma* species. Acta Mycologica 46(2): 179–186. https://doi.org/10.5586/am.2011.011

Tournefort JP (1700) Institutiones rei herbariae. Parisis, 697 pp.

White TJ, Bruns T, Lee L, Taylor JW (1990) Amplification and direct sequencing of fungal ribosomal RNA genes for phylogenetics. In: Innis MA, Gelfand DH, Sininski JJ, White J(Eds) PCR Protocols, A guide to Methods and Applications. Academic Press, New York, 315–322. https://doi.org/10.1016/b978-0-12-372180-8.50042-1

White VS (1901) The Tylostomaceae of North America. Bulletin of the Torrey Botanical Club 28: 421–444. https://doi.org/10.2307/2478590

Winterhoff W (2000) Epigäische Gasteromycetanae. In: Krieglsteiner GJ (Ed.) Die Grosspilze Baden-Württembergs Band 2: 103–183.

Wright JE (1955) Evaluation of specific characters in the genus *Tulostoma* Pers. Papers of the Michigan Academy of Science, Arts and Letters 40: 79–87.

Wright JE (1987) The genus *Tulostoma* (Gasteromycetes) – A world monograph. Bibliotheca Mycologica 113. Berlin, Stuttgart, 338 pp.

Wright JE, Herrera T, Guzmán G (1972) Estudios sobre el género *Tulostoma* in México (Fungi Gasteromycetes). Ciencia 27: 109–122.

Supplementary material 1

Unexpected high species diversity among European stalked puffballs – a contribution to the phylogeny and taxonomy of the genus *Tulostoma*

Authors: Mikael Jeppson, Alberto Altés, Gabriel Moreno, R. Henrik Nilsson, Yolanda Loarce, Alfredo de Bustos, Ellen Larsson

Data type: Table/specimen data

Explanation note: Data of specimens sequenced in this study with GenBank accession numbers for the ITS/LSU and TEF-1α regions. Type specimens are marked in bold.

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Supplementary material 2

Unexpected high species diversity among European stalked puffballs – a contribution to the phylogeny and taxonomy of the genus *Tulostoma*

Authors: Mikael Jeppson, Alberto Altés, Gabriel Moreno, R. Henrik Nilsson, Yolanda Loarce, Alfredo de Bustos, Ellen Larsson

Data type: Table/ sequence data

Explanation note: Distance matrices of the ITS alignment (uncorrected "p" distance and GTR distance) and the multiple sequence alignmnet of the ITS.

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